DEGRADATION OF BLEACHING AGENTS UNDER TWO DIFFERENT STORAGE CONDITIONS

by

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DEDICATION

To my loving parents,

To my wonderful wife,

To my little daughter, Sumo,

To all my family and friends,

For their endless love, support, and encouragement.

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INTRODUCTION

Tooth bleaching has become one of the most popular dental treatments in daily practice. Patients desire to have affordable, non-invasive cosmetic dentistry, performed safely, and in the shortest period of time, giving them a beautiful smile. At the same time, dentists want to provide treatment based on reliable evidence to determine the effectiveness and safety of various tooth bleaching delivery systems. Dentists are aware that effective tooth bleaching depends on the concentration of the active ingredient and its contact time with the tooth.

Manufacturing failures, storage conditions of the tooth-whitening agent, or shipping time to distant locations might be causative factors that make a difference between the labeled concentration and what the dentist receives. However, questions have also been raised about the degradation of the active bleaching ingredients once they arrive in the dental office. Chemical degradation of the active agent might occur if the dentist stores the product for an extended period of time or in warm conditions, causing the bleaching material to lose its potency.

The purpose of this study was to determine if there is any change in the active ingredient in the tooth-whitening agents when they are received from the manufacturer (Base-line), two months, four months, and six months after they are received, under 2 different storage conditions. The hypothesis is that the concentrations of bleaching agent at baseline, 2 months, 4 months, and 6 months are different when maintaining the active agents at room temperature or in a refrigerator.

REVIEW OF THE LITERATURE

TOOTH DISCOLORATION

Tooth discoloration is usually classified as extrinsic or intrinsic in nature depending on the place on which the chromogen is deposited. For extrinsic discoloration, external chromogens are deposited on the tooth or within the pellicle. Intrinsic discoloration occurs when the chromogens are deposited deep into dentine such as tetracycline stains or in the enamel such as flourosis.[1]

Tooth discoloration can be managed by different methods available in dental practice. Those methods range from whitening techniques to surgical removal of the underlying discoloration and restoring the tooth with direct resin, veneers or crowns.

Tooth discoloration has become one of the main reasons for patients to visit a dental office. According to a study conducted by Neumann et al.[2], patients were more concerned about the dental variables, including tooth color, than about orthodontic variables. In that study, it was suggested that the appearance of the teeth was considered a greater contributing factor to a cosmetic smile than was the position of the teeth.

Light can be reflected or absorbed by an object, which will affect the appearance of the object. The ability of the teeth to reflect or absorb the light is influenced by the components of the tooth, such as enamel and dentine. Any change that occurs to those components during formation, development, or after eruption can cause changes in the light transmission property and change the color of the teeth.[3]

Intrinsic Discoloration

Intrinsic discoloration is the most complicated type of tooth discoloration. It might occur during tooth development due to any change in the structural composition or thickness of the tooth component. It can be caused by multiple factors, such as metabolic, inherited, iatrogenic, traumatic, idiopathic, or aging factors.

Some metabolic disorders can cause teeth discoloration, such as alkaponuria, which results in a brown stain, or congenital erythropiotic porphyria, which is characterized by a red/purple–brown discoloration of the hard tissue of the tooth.[4, 5]

Amelogenesis imperfect is considered an inherited cause of tooth discoloration. It occurs when the mineralization of the enamel matrix is disrupted during tooth formation. The tooth becomes yellow-brown in color as an effect of that disruption.[6-8] The degree of hypomineralization is sometimes reflected in the tooth color; the lighter color the greater degree of hypomineralization. In addition, some types of dentinogenisis imperfect are also considered as an inherited cause of the internal tooth discoloration. The affected teeth tend to be amber or gray to purple-blue in color.[9]

Tetracycline staining occurs as a result of administration of the tetracycline antibiotic, one of the wide spectrum antibiotics.[10] It should be avoided during formation of the teeth from the second trimester until the child is eight years old.[11] The appearance of the teeth vary from yellow to brown-gray in color.[12] In 1984 Jordan and Boshman[13] divided the tetracycline discoloration into three major categories according to the extent, degree, and location of tetracycline involvement:

- Degree I: Light yellow stains uniformly confined to the incisal three-quarters of the crown without color band.

- Degree II: Highly uniform deep yellow to gray stains with no banding.
- Degree III: Very dark uniform blue or gray stains with banding.

Fluorosis is one of the most common intrinsic discolorations seen in dental clinics. It occurs as a result of ingesting excessive fluoride from one of several different sources, such as fluoride in water supplies, mouth rinse, or toothpaste. The critical period, in which there is a greater probability for fluorosis to occur, is from the third month of pregnancy through the eighth year of life.[14-16] This kind of discoloration mostly appears as a hypocalcification in the enamel of the tooth. Some brown discoloration is caused by internalization of external stains into the porous enamel.

Trauma to the tooth is also considered as a common cause of tooth discoloration seen in dental offices. The tooth might look pink in the early days after the trauma due to bleeding inside the pulp.[17] Later, the hemoglobin in the blood degrades to produce some iron. This iron laterally combines with the hydrogen sulfide component inside the dentinal tubule to produce iron sulfide. Iron sulfide will give the tooth a gray-blue or black appearance.[16, 18-20] Deposition of hard tissue within the root canal space might occur following trauma, known as calcific metamorphosis. Complete or partial pulp chamber obliteration may cause the clinical crown to look yellow.[21] Trauma to primary teeth can lead to localized enamel hypoplasia in successor permanent teeth.[22]

Aging also causes the darkening of teeth. If the enamel gets thinner or there are changes in texture, the light transmission properties of the tooth will be affected. In addition, the depositions of secondary and tertiary dentin play an important role in darkening the tooth with age.[23]

Some of the endodontic and restorative material can also cause changes in the

tooth color. These include silver cone or silver-containing sealer, which are no longer used in modern dentistry; silver amalgam, which might cause a gray-black discoloration due to migration of tin into the dentinal tubule; and a eugenol-containing restoration, which might cause an orange-yellow stain.[16]

Extrinsic Discoloration

Extrinsic discoloration can be caused by direct adherence of the stain to the outer surface of the tooth with no chemical changes in the enamel. It can also be caused by deposition of plaque and calculus onto the tooth surface.[24]

The most common causes of the extrinsic discoloration are coffee, tea, artificial food colors, some type of fruits, and smoking.[25] The deposition of these stains mainly occurs in the gingival margin and interproximal area of the tooth, where it is more difficult to maintain oral hygiene.

Dental caries lesions are also considered as a cause of the extrinsic discoloration, the color ranging from white spots to brown and black lesions, which get stained from external sources.[26]

BLEACHING CHEMISTRY

Depending on the environmental conditions, such as temperature, pH, ultraviolet (UV) light and the presence of some ions, the hydrogen peroxide (H_2O_2) breaks down into water (H_2O) and an oxygen free radical. The hydrogen peroxide acts as an oxidizing agent through the formation of free radicals, reactive oxygen molecule ($O^{\bullet}$) and hydrogen peroxide anion ($HO_2^{\bullet}$).[27, 28]

$$H_2O_2 \leftrightarrow 2HO\bullet$$

$$HO\bullet + H_2O_2 \rightarrow H_2O + HO_2\bullet$$

$$HO_2\bullet \leftrightarrow H^+ + O_2\bullet$$

In 1991 Albers[29] described the more extensive bleaching reaction. Carbon-ring compounds of complex organic molecules with high molecular weight pigments are broken down by the chemical reactions with the free radicals to form long chains of carbon double-bond compounds. Then, the carbon double-bonds degrade by the action of the free radicals into a simpler molecule with hydroxyl group ends, which absorb less light and are essentially colorless. As a consequence, the change in the molecule size leads to a change in the light reflection and the change of the stain to a lighter color.[29-31]

Perhydroxyl ($HO_2\bullet$) is a stronger and more reactive free radical. Hydrogen peroxide (H_2O_2) needs to be made in a high pH environment to promote formation of a greater amount of the Perhydroxyl ($HO_2\bullet$) free radical as shown in the following reaction:[32, 33]

$$H_2O_2 \leftrightarrow H^+ + HOO\bullet$$

A free radical will not be produced in the presence of decomposition catalyst and enzymes. In this situation, the hydrogen peroxide reaction will occur as follows:[34]

$$2H_2O_2 \leftrightarrow 2H_2O + 2\{O\} \leftrightarrow 2H_2O + O_2$$

The carbamide peroxide ($CH_6N_2O_3$), also called urea peroxide, contains hydrogen peroxide loosely associated with urea. Ten percent of carbamide peroxide breaks down into 3.6% hydrogen peroxide and 5.4% urea, which also decomposes to carbon dioxide and ammonia. Ammonia can elevate the pH of the reaction to positively facilitate the bleaching process. The chemistry of carbamide peroxide is slightly different from

hydrogen peroxide. The presence of urea will make the degradation slower than hydrogen peroxide alone, which permits the peroxide to remain in contact with the tooth for a longer period of time.[35-38]

Sodium perborate ($NaBO_3$) is the material of choice to perform internal bleaching. It is stable when in dry powder form. During the bleaching reaction it decomposes in the presence of acid, moisture, or water into metaborate, hydrogen peroxide and nascent oxygen, as shown in the following reaction:[39, 40]

$$Na_2\,[B_2(O_2)_2\,(OH)_4] \quad + \quad 2H_2O \quad \rightarrow \quad 2Na\,Bo_3 \quad + \quad 2H_2O_2$$

NON-VITAL BLEACHING

There are three non-vital bleaching techniques available. These are: walking bleaching, non-vital power bleaching, and inside/outside bleaching.

Walking Bleaching Technique

In 1859 Spasser[41, 42] was the first to describe walking bleaching in the dental literature. A mixture of sodium perborate and water were sealed into the pulp chamber of the tooth. This procedure would be repeated every week until the desired color of the tooth had occurred. There is also another technique called modified or combination walking bleaching. In this technique, a combination of 30% hydrogen peroxide and sodium perborate are sealed into the pulp chamber of the tooth for one week.[39, 43] When hydrogen peroxide is mixed with sodium perborate, it forms a thick paste and leads to an increased bleaching effect.[40]

Today, 10% carbamide peroxide is sometimes used instead of a mixture of sodium perborate and hydrogen peroxide. Carbamide peroxide is simply sealed into the pulp chamber of the patient and followed up every 3 days until the desired color has been achieved.[43]

Non-Vital Power Bleaching

In this technique, thirty to thirty-five percent hydrogen peroxide is applied into the pulp chamber and activated with either heat or light. After that, the temperature is increased gradually up to 50-60°C for 5 minutes.[40] The walking bleaching technique is used between visits until the tooth gets the favorable whitening.[42]

In a variation to this technique, 35% hydrogen peroxide gel is applied in the pulp chamber and on the facial surface of the tooth. Then, light activation is used for both internally and externally placed gel.[44]

Inside/Outside Bleaching

A combination of internal bleaching of the non-vital bleaching tooth and at-home bleaching are required in this technique. It is the most favorable technique for bleaching a non-vital tooth because a lower concentration of bleach can be used, usually 10% carbamide peroxide, to reduce the risk of external resorption of the root. This technique gives a better tooth whitening result in less time in comparison to other non-vital bleaching techniques. [44]

VITAL BLEACHING

In-Office Bleaching

Vital bleaching was introduced to dental practices as early as 1868. It was done by using oxalic acid first and later using hydrogen peroxide.[45] In 1911 Dr. Roseunthal[46] suggested including the use of violet and ultraviolet light in combination with hydrogen peroxide to improve the result of the teeth bleaching.

In 1970 Cohen and Parkins[47] reported that using 30% hydrogen peroxide alone showed a successful result when it was warmed by a controlled device to about 88°F. This technique was done in eight separate appointments with a 30-minute application at each visit.

By the beginning of the 1980s, removing enamel stains was well documented and included removing fluorosis discoloration. However, some problems were related to removal of dentin staining, such as in tetracycline cases. In 1982 Murrin and Barkmeier[48] attempted to introduce a new approach to treat tetracycline and other intrinsic discoloration. They recommended applying 36% hydrochloric acid for one to two minutes as an acid etchant prior to the bleaching. Feinman et al.[16] introduced a similar technique by using a phosphoric acid for 20 seconds as an etchant prior to applying 35% hydrogen peroxide. A high-intensity light was used for 30 minutes to keep teeth warmed during the application of hydrogen peroxide. Depending on the discoloration type and severity, multiple treatment visits were needed to achieve the best bleaching result.

By the beginning of the 1990s, techniques for vital bleaching had been improved and they started to be called “In-Office” or “Power” bleaching. The teeth were pre-etched

with phosphoric acid before applying a 35% hydrogen peroxide bleaching agent. A heated instrument, instead of a light source, was used to enhance the action of the peroxide.[15, 16, 29, 49] The treatment generally took place for 30-45 minutes during approximately four to six visits. Patients were not anesthetized in order to allow them to determine the proper heat level. Isolation of the heat and surrounding tissue were mandatory in order to eliminate direct contact of the caustic bleaching gel with gum, cheek and tongue.

Some studies evaluated the effect of the heat and hydrogen peroxide concentration on the pulp. Those studies showed that the pulp remained vital and the damage is reversible in about two months.[50-53]

In 1985 Hall[52] reported a study conducted to evaluate the effect of pre-etching teeth prior to performing bleaching. He reported that there was no advantage to pre-etching teeth with phosphoric acid in preparation for bleaching. In addition, Papathanasiou et al.[54] conducted a study to compare the effectiveness of light activation and no light activation of 35% hydrogen peroxide. They concluded that there was no benefit to using light activation over no light activation. In another study by Hein et al.,[55] it was concluded that neither the light nor the heat produced by the light had any benefit on the efficiency of In-Office bleaching. On the other hand, Dostolova et al.[56] showed that using a diode laser combined with 38% hydrogen peroxide decreased the time of In-Office bleaching.

In recent research, Sulieman et al.[57] showed that most bleaching lamps increase the interpulpal temperature less than 5.5°C, which is the critical threshold of producing irreversible damage to the pulp. The laser-based lamp is the only lamp that produces an

interpulpal temperature above 5.5° C. To overcome this problem, the power output of the laser-based lamp intensity should be reduced to 2 W.

As to the advantages of In-Office bleaching, it has been found to be the proper technique for those patients who have no time available for home bleaching, who have a problem with wearing trays, or who need some motivation to continue with At-Home bleaching.[44]

At-Home Bleaching

At-Home bleaching became very popular right after it was introduced by the dental profession. It involves application of bleaching gel by the patient into a bleaching tray. A vacuum-formed, soft plastic custom tray is fabricated by the practitioner and given to the patient along with the manufacturer's instructions. This technique is also called night-guard bleaching.[58] It is the most favorable technique since it is less dangerous and has fewer side effects, along with a lower cost.

In 1968 Dr. Bill Klusmier, an orthodontist in Little Rock, AR, noticed that mild tetracycline stains become lighter after an extended usage of Gly-Oxide, which was used to treat inflammation problems during orthodontic treatment. He presented his findings in multiple meetings and started to use Gly-Oxide after each orthodontic treatment. After that, he switched to another oral antiseptic gel called Proxigel because he found that it was more viscous and stayed in the appliance longer.[59]

In 1986, using Proxigel in a plastic night-guard became widely used by the Coastal Dental Study Club. In the same year, Dr. John Munro, a general dentist from Tennessee, reported that using 10% carbamide peroxide in a vacuum-formed plastic tray

for 3 to 7 days could change the teeth to a whiter color. He noted that when peroxide oxygenation was used to control the bacterial growth after periodontal root planning, teeth became lighter in color.

Manufacturers, in 1989, started to develop 10% carbamide peroxide gel to be used as a daytime-use bleaching product. One year before that, Dr. Van Haywood and Dr. Harold Heyman began clinical trials at the University of North Carolina to study the efficiency of At-Home bleaching.[59] As a result of these trials, they published the first report about At-Home bleaching, using 10% carbamide peroxide in a custom tray worn at night.[60]

By the early twenty first century, there were several bleaching products available on the market. Most of those products have been shown to work to a comparable extent.[61]

The advantages of At-Home bleaching include ease of application, reduced chair-time, and reduced cost, making it a favorable technique for both patients and dentists. In addition, the high percentage of success, the safety of materials, and the elimination of heat or etching are considered advantages of using At-Home bleaching.[44]

EFFICINCY OF BLEACHING ACTIVE AGENT

In 2004 Sulieman[62] et al. studied the effect of 35% hydrogen peroxide on tea staining in comparison to distilled water. They concluded that hydrogen peroxide had the bleaching efficiency to remove stains. In 2005 the same authors compared the effect of various concentrations of hydrogen peroxide on the outcome of tooth whitening. They demonstrated that, to obtain a specific whitening outcome, a higher concentration of hydrogen peroxide required fewer applications.[37]

Zekonis et al.[63] compared the color changes associated with a highly concentrated in-office bleaching compared to a lower concentrated at-home bleaching. They showed that 14 days of at-home bleaching was more effective than two applications of in-office bleaching. However, Auschill et al.[64] reported that all tooth bleaching techniques were effective in teeth whitening with special consideration to the contact time required to achieve the favorable whitening.

Leonard et al.[36] compared the efficiency of three different concentrations of carbamide peroxide: 5%, 10%, and 16% of the same product. They reported that, after 2 weeks of the teeth whitening, both 10% and 16% carbamide peroxide showed no difference. However, after 3 weeks of whitening, all 3 concentrations gave comparable results. They concluded that the higher concentration of carbamide peroxide provided more rapid lightening compared to the lower concentrations. Another study, done by Matis et al.,[65] showed that, after 2 weeks of bleaching, a 15 % concentration resulted in a faster and greater teeth whitening than a 10 % concentration. However, 4 weeks post-bleaching evaluation showed no statistically significant difference between all concentrations. In both the Leonard et al.[36] and Matis et al.[65], it was concluded that the lower-concentration carbamide peroxide gave equal lightening when it was applied for a longer period of time.

Panich [30] and Mokhlis et al.[66] conducted studies to compare the effectiveness of hydrogen peroxide to carbamide peroxide in teeth whitening. Panich[30] used an equal amount of active agents for both hydrogen peroxide and carbamide peroxide, while Mokhlis[66] used a higher concentration of hydrogen peroxide and carbamide peroxide, which had close to the same amount of active agent. Both studies concluded that an equal

concentration of hydrogen peroxide, whether alone or in carbamide peroxide, produces a similar whitening effect with equal contact time.

In general, a more lightening effect can be achieved by using higher-concentration products and more viscous and thicker materials. However, using the higher concentration products could lead to a greater chance of thermal sensitivity during and after the treatment.[67] Therefore, selection of a favorable concentration depends on multiple factors, including how dark is the tooth; the type of discoloration; the patient's lifestyle and preferences; and, the most important factor, the teeth sensitivity during bleaching procedures.[44]

DEGRADATION OF BLEACHING AGENT

The agent in the bleaching gel needs to be active for an extended period of time for the bleaching process to occur. Many studies have been done to examine how long the bleaching agents remain active once they are placed on the teeth.[68-74] One clinical study done in 1997 [70] found that less than 50% of the active bleaching agent was present in nine popular bleaching products after half an hour of mouth application. Another clinical study was done in 1999 by Matis et al.[69] to determine the in vivo degradation rate of 10% carbamide peroxide gel in the bleaching tray. The bleach concentrations were determined for six time intervals ranging from 15 seconds to 10 hours. They reported that the average degradation of carbamide peroxide in the first hour was 63%. In the same year, a study was done by Wattanapayungkul et al.[71] to determine the effect of pellicle on that degradation. They concluded that the degradation rate was not affected by the pellicle removal. In 2002 Dr. Matis et al.[74] conducted an in vivo study to determine the effect of

tray design on the degradation of 9 carbamide peroxide products. They found that the whitening gel in both trays, with or without a reservoir, degraded at the same rate after 2 hours of use. One year later, Alqunaian et al.[69] conducted a study similar to Wattanapayungkul's [71] study, but their study was done to determine the kinetics of 3% hydrogen peroxide in bleaching gel within the first hour. They reported that the average remaining hydrogen peroxide was 32.23% at the end of one hour of clinical application and that the degradation was higher in the first 10 minutes. In 2013 Alonso De La Pena et al.[75] conducted an in-vivo study to determine the degradation of two At-Home bleaching agents, one hydrogen peroxide and one carbamide peroxide of similar H_2O_2 concentration. They compared the degradation of the active ingredient in the tray intraorally at different time intervals ranging from 5 minutes to 75 minutes. More than 50% reduction in carbamide peroxide concentration was recorded after 40 minutes, while 50% of hydrogen peroxide declined after 60 minutes of application. They also found that the concentration of the active ingredient was 8.12% in the hydrogen peroxide product and 7.95% in the carbamide peroxide product, which was higher than those given by manufacturers. In all previous degradation studies, at-home bleaching was used and showed intense degradation a few hours after their application.

Tooth bleaching has been researched extensively from different points of view. All of these studies were based on the labeled concentration of the active bleaching agent, which might not be the actual concentration. Multiple studies have concluded that there are discrepancies between the labeled and the actual concentration of the active agent in the bleaching material.[73, 76, 77] In 2013 Matis et al.[77] compared the actual and labeled concentration of different available bleaching products in four different parts of the

world. The products were tested when they were received and, for the United States' samples, during the month just before expiration. For many products, the actual concentration of the active agent was different from that listed on the label. Most of them were within the required range set by the International Organization of Standardization[78] (ISO) (10% higher to 30% lower than concentration listed on the label) when tested upon receiving the product. One product in United States, five products in Saudi Arabia and six products in Brazil had a loss of more than 15% of the concentration indicated on the label. However, one product in Saudi Arabia and three products in Brazil had a loss of more than 30% of the concentration indicated on the label. Another study by Martin et al.[73] was conducted in Brazil to analyze the concentration of 16% carbamide peroxide in 100 samples of 4 compounding pharmacies, and one commercially available bleaching gel was used as a control group. They found that the control group presented a closer mean concentration value to 16%. However, concentrations of both manipulated and industrialized carbamide peroxide presented concentration values different from what was stated on the label, ranging from 8 % to 20 %. This means that, in bleaching studies and while treating patients, researchers and dental practitioners are not always starting with the concentration indicated on the label by the manufacturer.

In one of the previously mentioned studies, Matis et al.[74], stated that after completion of the study, evaluation of the bleaching agent showed a slight decrease in value compared to the baseline.[74] That might be caused by storing the bleaching products for an extended period of time while the study was being conducted.

STORAGE RECOMMENDATION

Temperature is the most critical the environmental factors that contribute to drug degradation.[79] Manufacturers must guarantee a potency of 90% to 110% for medical drugs.[80] Stability tests performed under different temperatures for a specific period were done for any drug to determine the expiration date and storage conditions that should be recommended. The manufacturer should guarantee the stability of drugs if it is stored as recommended until the expiration date.[81]

One recent longitudinal study was done to evaluate the stability of drugs requiring refrigeration under different temperature exposure.[82] They compared the stability of five drugs stored for one year under three storage conditions; in the refrigerator at 2°C (35.6°F) to 8°C (46.4°F), at room temperature (20°C to 25°C) (86°F to 77°F) and in emergency physician transport vehicles. As a result of their study, they found that one drug became unstable within weeks, two drugs become unstable within months and the others stayed stable for several months.

As for bleaching products, the stability is a concern to dentists and researchers. Since the storage temperature might affect the concentration of the active ingredient and the efficiency of tooth whitening. Some manufacturers recommend that their bleaching products be stored in a refrigerator, while the others recommend storing their bleaching products at room temperature. Although some manufacturers recommend that dentists refrigerate whitening products once received, manufacturers do not refrigerate all whitening products during storage or the shipping process. Only one manufacturer (KöR) sends their product in a refrigerated pack (Figure 11) because they are concerned about loss of concentrations during the shipment. They claimed that refrigeration should be

performed from the instant of manufacture until the dental office receives the product to stop the degradation of peroxide in bleaching gels to lengthen shelf life.[83]

Only one study has been performed to study the effect of storage temperature on the bleaching agents. Freire A. et al.[84] conducted a study aimed to determine the pH of several commercially available In-Office and At-Home dental bleaching products stored at two different temperatures, room temperature (23°C±1°C) and refrigeration temperature (4°C±1°C). That study demonstrated that lower pH values were found when the products were stored at room temperature.

To date, to our knowledge, there is a lack of studies investigating the effect of long-term storage conditions on the active concentrations of bleaching products with and without refrigeration. This research study is to determine if there is any change in the concentration of the active agent in the tooth-whitening agents when they are received from the manufacturer (Base-line), two months, four months, and six months after they are received, under 2 different storage conditions.

METHODS AND MATERIALS

PROCEDURES AND METHODS

Manufacturers of tooth-whitening agents available in United States were requested to forward two samples of each of their products to IUSD. Eleven manufacturers of whitening agents each forwarded two of their At-Home bleaching products of various concentrations. Thirty-six products were received: eight hydrogen peroxide and 28 carbamide peroxide products (Table I). Manufacturers provided different concentrations and appropriate storage instructions (Table II). All the bleaching syringes for a specific product were from the same lot. Once the products were received, one sample of each product was stored in a cabinet at room temperature and the other sample was stored in a refrigerator.

Room temperature was monitored in the morning during the period of storing the products. The room temperature was within the range from 20.3°C (68.6°F) to 22.78°C (73°F). The refrigerator temperature was held constant at 5°C (41°F).

The first product was received at the end of November while the last product was received in the middle of January. Assays to determine the baseline concentration were performed within the first two weeks of the products' arrival. In addition, assays were performed 2 months, 4 months and 6 months after receiving the products. One to two bleaching syringes, from the same lot number, were used to complete the study for each product in each storage condition. The protocol, recommended by the United States

Pharmacopeia and ISO Standard[78], was used for determining the amount and concentration of peroxide in the tooth-whitening agents.

CHEMICAL ANALYSIS OF BLEACHING AGENT

Preparation

Prior to determining the amount of peroxide in a bleaching agent, the current date, manufacturer, product, expiration date, peroxide type, peroxide concentration, and trial number were recorded on a working sheet. Data recording sheets are shown in Appendixes A and B.

Approximately 0.2 g of the bleaching gel was weighed on top of weighing paper, which was tared on the analytical balance (Mettler AE100, ±0.1 mg, Mettler Toledo, Columbus, OH) (Figure 1). Deionized water (Milli-Q® Plus Water System, Millipore Corp., Bedford, MA) was used to wash out the gel from weighing paper into an empty 400-ml beaker using a wash bottle (Figure 2). The pH of the deionized water was measured prior to every assay performed. It was within the range of 5.67 to 5.95. Then, the beaker was filled up to the scale of 100 ml with Milli-Q water. A stir bar was added, and the beaker contents were mixed on a stir plate (Thermix® Stirrer, model 220t, Thermo Fisher Scientific, Waltham, MA) until a homogeneous mixture was attained (Figure 3). Twenty milliliters of glacial acetic acid (Glacial acetic acid certified ACS, Fisher Scientific, Fair Lawn, NJ) was added (8 ml of glacial acetic acid per each 40 ml of the beaker contents) and the beaker was immediately covered using a glass dish (Figure 4). Then, the solution was stirred for approximately five minutes or until the gel was

completely dissolved. About two gram of potassium iodide (Potassium iodide certified ACS, Fisher Scientific, Fair Lawn, NJ) was weighed on the balance scale (Fisher balance Model S-400, Thermo Fisher Scientific, Waltham, MA) and added to the solution. This turned the solution to a light shade of yellow (Figure 5). Three drops of ammonium molybdate (Ammonium molybdate 4% (W/V) Aqueous Solution ACS Grade, Ricca Chemical Co., Batesville, IN) were then added using a disposable plastic pipette, and the solution was again allowed to become homogenous. The solution turned to a dark shade of yellow or orange after adding ammonium molybdate (Figure 6). The beaker was then placed in a dark area for at least10 minutes. This time in the dark allowed the chemicals to fully associate to ensure a complete reaction with the available peroxide agent.

Titration

The beaker was then placed on the stir plate and the first titration started (Figure 7). Gradually, 0.01 N sodium thiosulfate (Sodium thiosulfate VS 0.01 N Cert, Thermo Fisher Scientific, Fair Lawn, NJ) was titrated into the solution, using a 50-ml burette, until the sample turned to a pale shade of yellow (Figure 8). The first titration volume was recorded. Then, three milliliters of a 1.0% starch indicator (Starch Indicator 1% Aqueous solution, Aqua Solution, INC., Deer Park, TX) was added to the solution, turning the solution a dark purple (Figure 9). More sodium thiosulfate was titrated into the solution as a second titration using 0.025 N sodium thiosulfate (Sodium thiosulfate VS 0.025 N Cert, Fisher Scientific, Fair Lawn, NJ) in a 10-ml burette, until the solution became colorless, which was the end point of the assay (Figure 10). All chemical analyses of concentrations were performed in triplicate.

Chemical Reaction

Hydrogen peroxide oxidizes potassium iodide to iodine in the presence of acetic acid and ammonium molybdate catalyst. This reaction is actually an oxidation/reduction reaction. The acetic acid is being used to provide an acidic environment for the oxidation-reduction reaction to take place in. The peroxide value is determined by measuring the amount of iodine, which is formed by the reaction of peroxides with iodide ion.
The iodine formed is titrated with sodium thiosulfate solution, incorporating a starch indicator. [85]

Calculation

The following formulas were used to calculate the peroxide concentration (wt %) depending on the type of peroxide in the sample, hydrogen peroxide or carbamide peroxide.

Hydrogen Peroxide (HP%) = V N (1**.**704)/ W

Or

Carbamide Peroxide (CP%) = V N (4**.**704)/ W

V= volume of sodium thiosulfate (ml).

W= weight of sample (g).

N= the normality of sodium thiosulfate.

Statistical Analysis

The triplicate analyses to determine the concentration for each sample were averaged before conducting the statistical analyses. The relative degradation of each

product was assessed using the baseline concentration–(measured concentration-baseline concentration) / baseline concentration * 100%. One-sample t-tests were used to test for a significant reduction in concentration for each storage method-time combination. Linear mixed effects models were used to evaluate the effects of time, storage method, and the time-storage interaction on relative degradation.

RESULTS

Thirty-six products were enrolled in this study, eight with hydrogen peroxide and 28 with carbamide peroxide. The hydrogen peroxide concentrations ranged from 6% to 15%. The carbamide peroxide concentrations ranged from 10% to 35%. The minimum concentration was 6%, and the maximum concentration was 35% (Table I). The 432 assays were performed during the study. Tables III-X show the mean concentration of the three tests done to determine the actual concentration for each product at baseline, 2 months, 4 months and 6 months, respectively.

The percentages of peroxide concentration change for each time interval, in both room temperature samples and refrigerated samples, are shown in Table XI. The means of the relative change in concentration for the refrigerated samples, using the label concentration as the reference, were -4.3 ± 5.56 at baseline. The means of the relative change in concentration for the room temperature samples, using the label concentration as the reference, were -5.35 ± 5.40 at base line. The means of the relative change in concentration for the refrigerated samples, using the baseline concentration as the reference, were -1.60 ± 1.76 at 2 months, -3.09 ± 3.30 for 4 months, and 2.85 ± 2.30 for 6 months. The means of the relative change in concentration for the room temperature samples, using the baseline concentration as the reference, were -3.50 ± 2.50 at 2 months, -4.41 ± 4.04 at 4 months, and -4.87 ± 4.02 at 6 months.

Table XII shows the mean (95% CI) for relative change in concentration. There was a significant decrease in concentration compared to the baseline concentration for each storage-time combination ($p \leq 0.0001$) (Figures 11 and 12).

No significant interaction was found between time and storage method ($p=0.43$ for analyses using baseline as the reference). The storage method had a significant effect on relative degradation, with more degradation for room storage than for refrigerated storage ($p \leq 0.0001$).

Using the label concentration as the reference, baseline had less degradation than 2 month ($p<0.0001$). Using the baseline concentration as the reference, 2 month had less degradation than 4 month ($p=0.0001$) and 6 month ($p=0.0001$) but 4 month and 6 month were not significantly different from each other ($p=0.81$).

Thirty-four products were within 15% of the active agent indicated by the manufacturers at base line. Two products had a 15% lower concentration of active agent but not more than 30% of that indicated by manufacturers (Tables III and VII).

For the room temperature samples: four products for the two-month assays, nine products for the four-month assays and six products for the six-month assays, were lower than 15% concentrations of what was stated in the label in the two-month assays (Tables IV, V, VI). One product had more than 30% lower concentration of active agent than what was on the label in the six-month assays.

For the refrigerated samples: three products for the two-month assays, five products for the four-month assays, and three products for the six-month assays were lower than 15% concentrations of what was stated on the label in the two-month assays

(Tables VIII, IX, X). All the products were not more than 30% of the concentration indicated by the manufacturers.

FIGURES AND TABLE

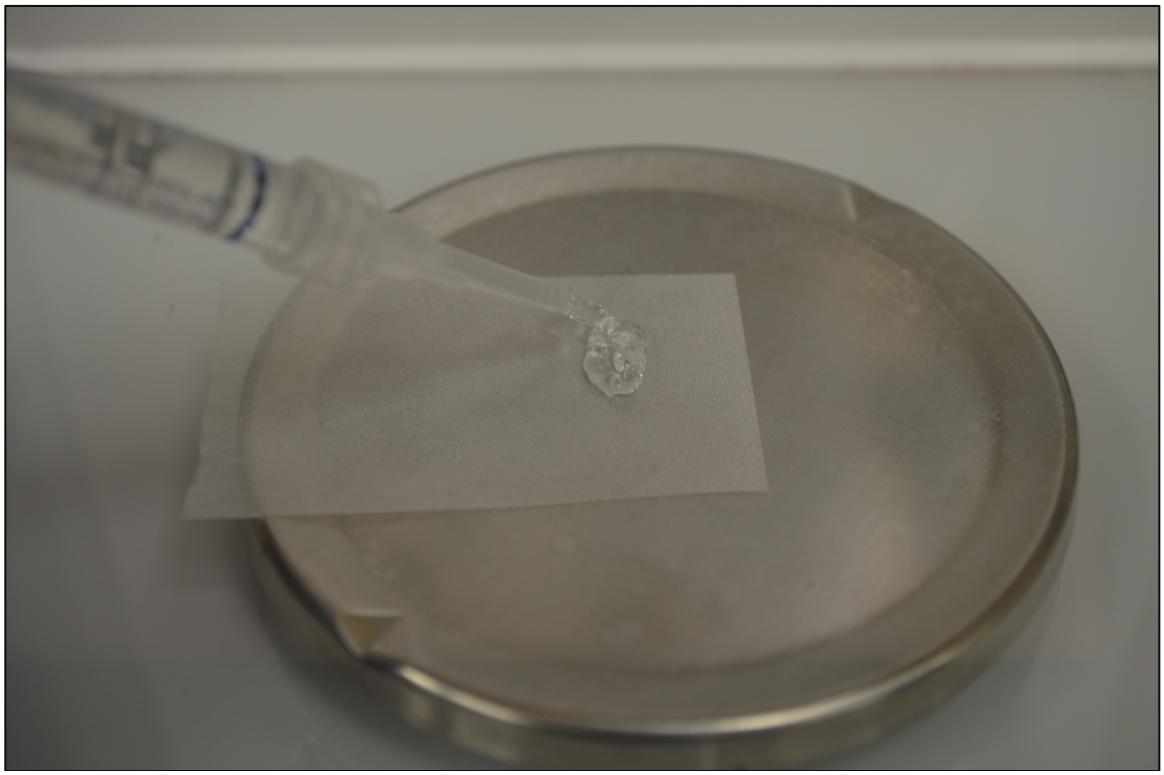

FIGURE 1: Weighing bleaching gel on the analytical balance.

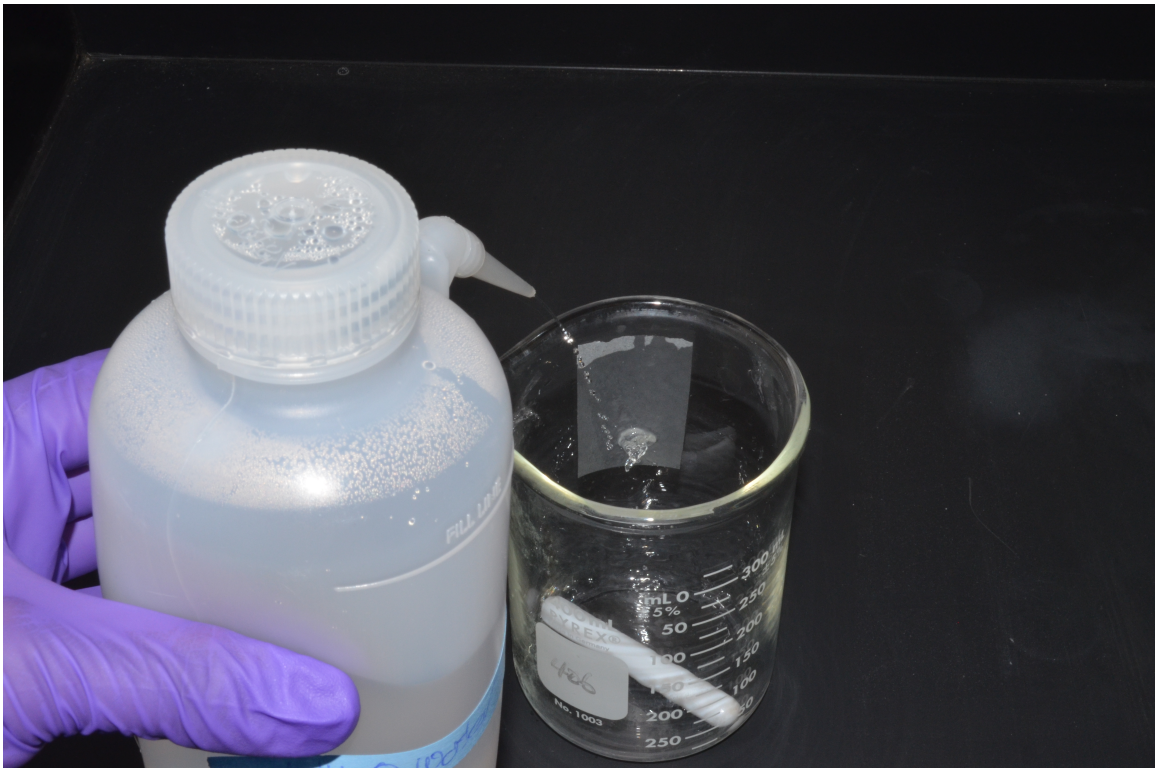

FIGURE 2: Washing out the bleaching gel into the beaker.

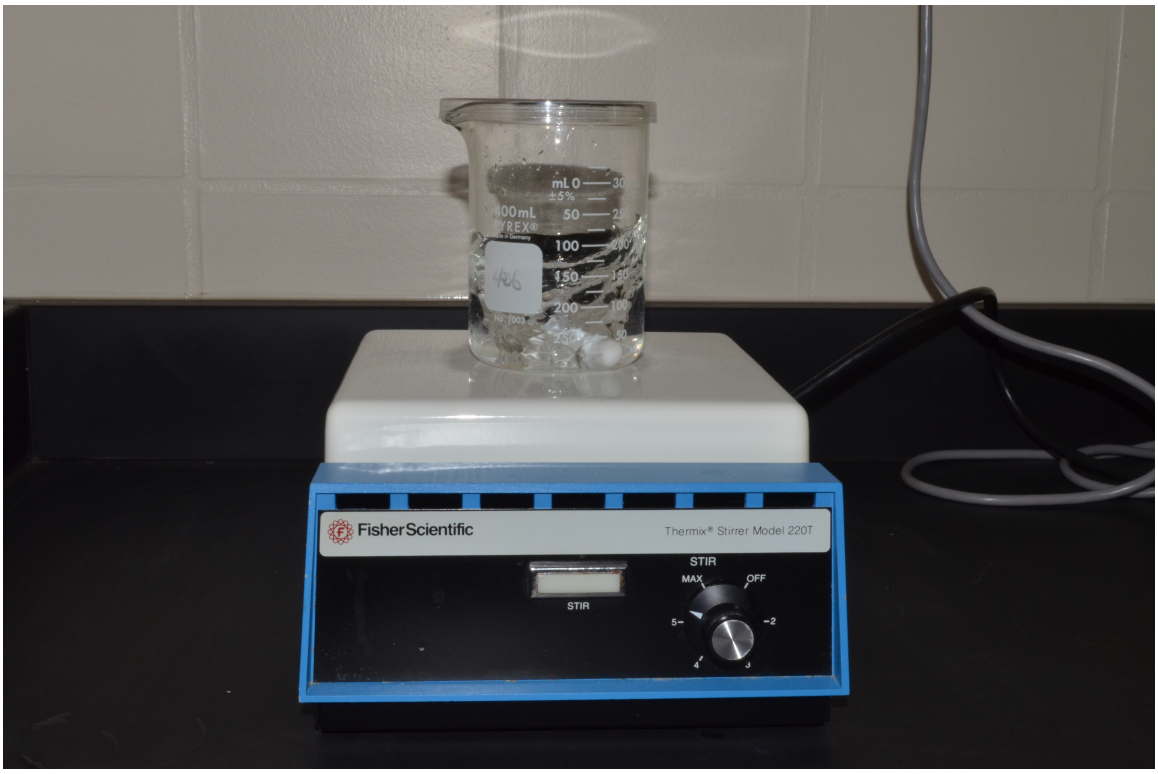

FIGURE 3: Beaker containing the bleaching gel and deionized water mixed on a stir plate.

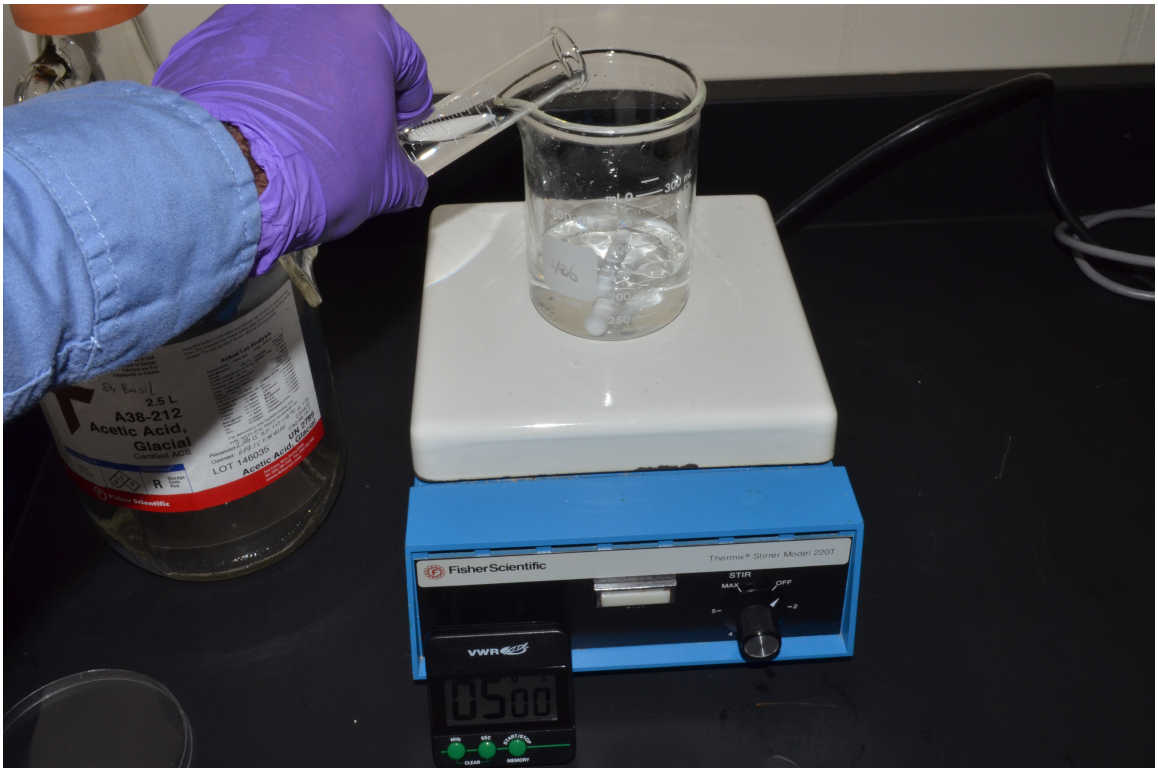

FIGURE 4: Adding glacial acetic acid to the solution.

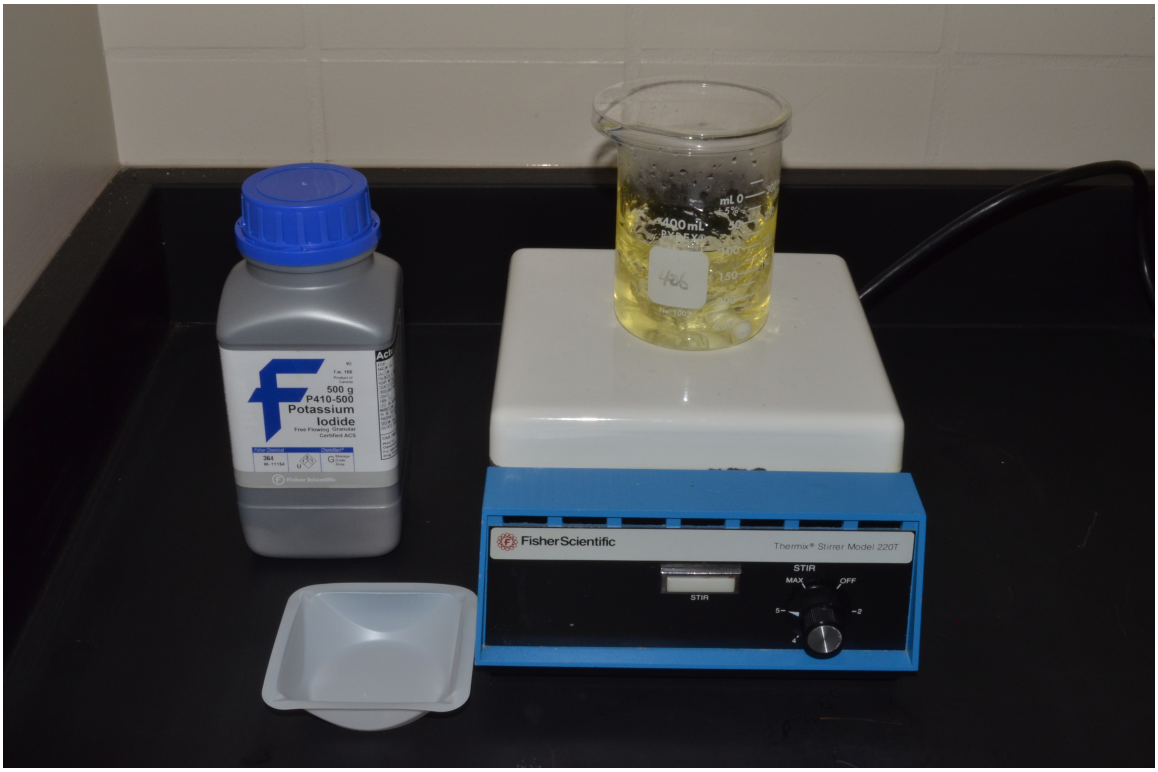

FIGURE 5: Potassium iodide turned the solution to a light shade of yellow

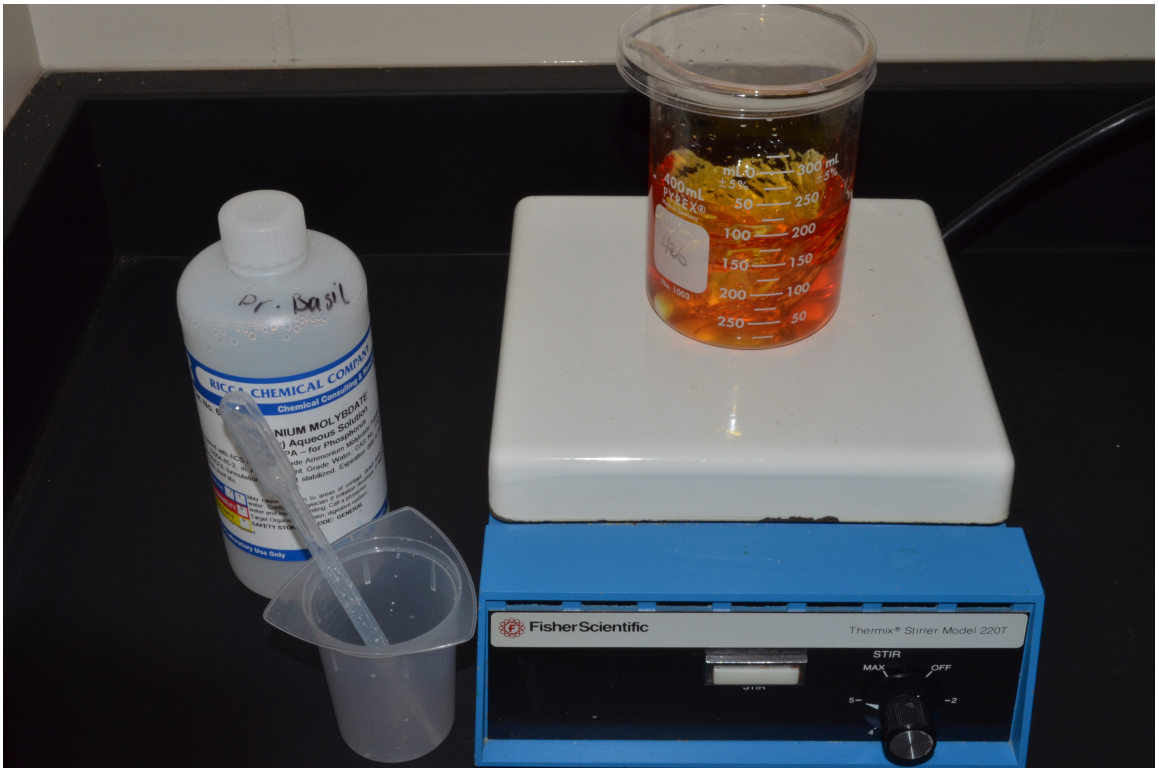

FIGURE 6: The solution turned to a dark shade of yellow or orange after adding Ammonium molybdate.

FIGURE 7: The beaker placed on the stir plate for the first titration.

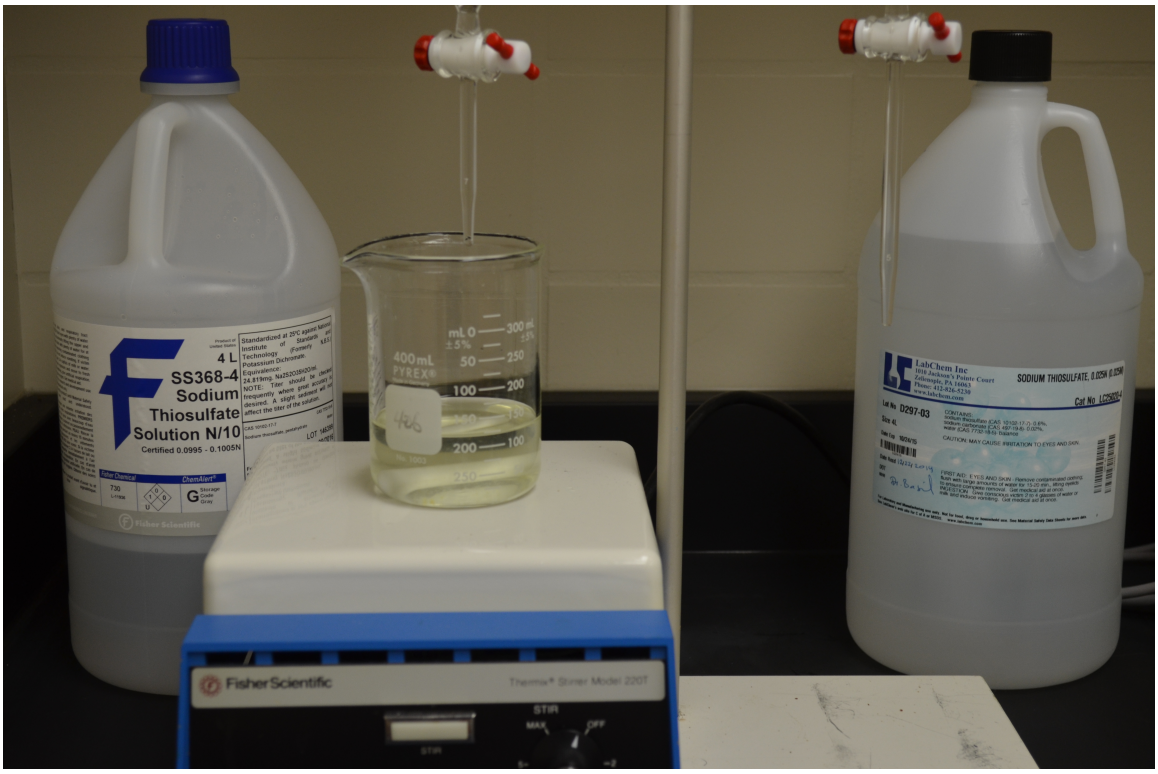

FIGURE 8: The beaker placed on the stir plate for the second titration.

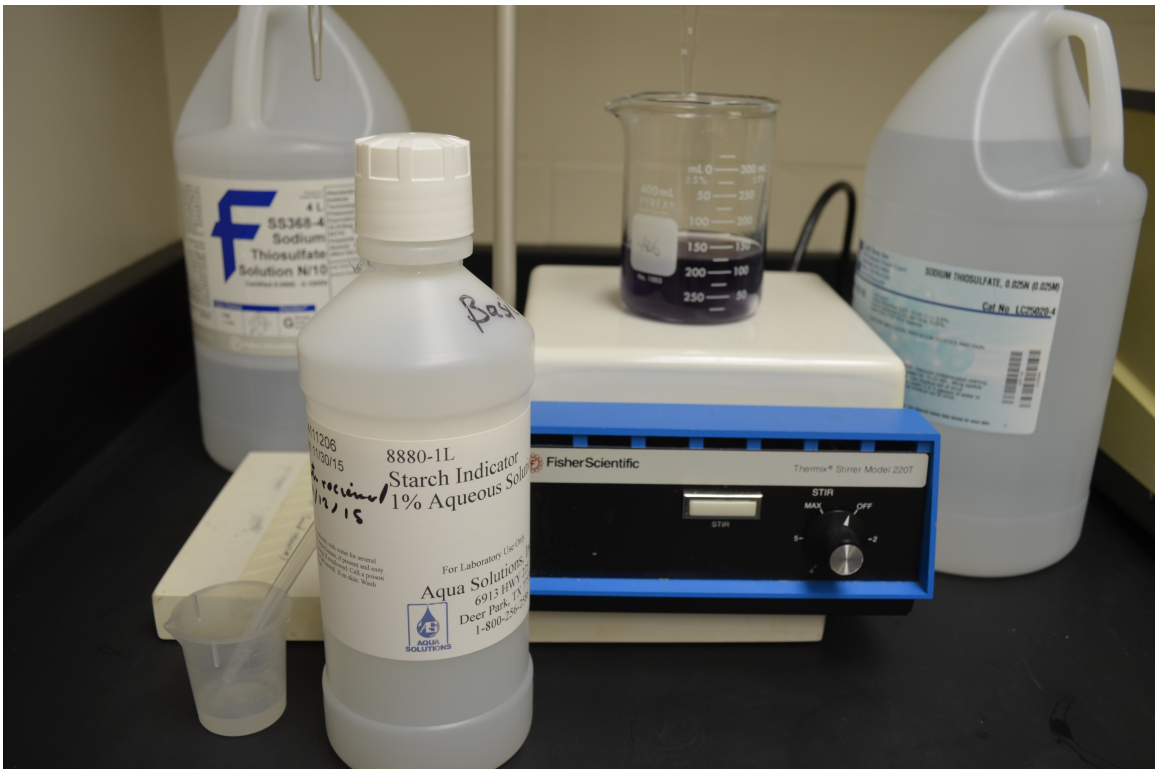

FIGURE 9: The solution becomes purple after adding 1.0% starch.

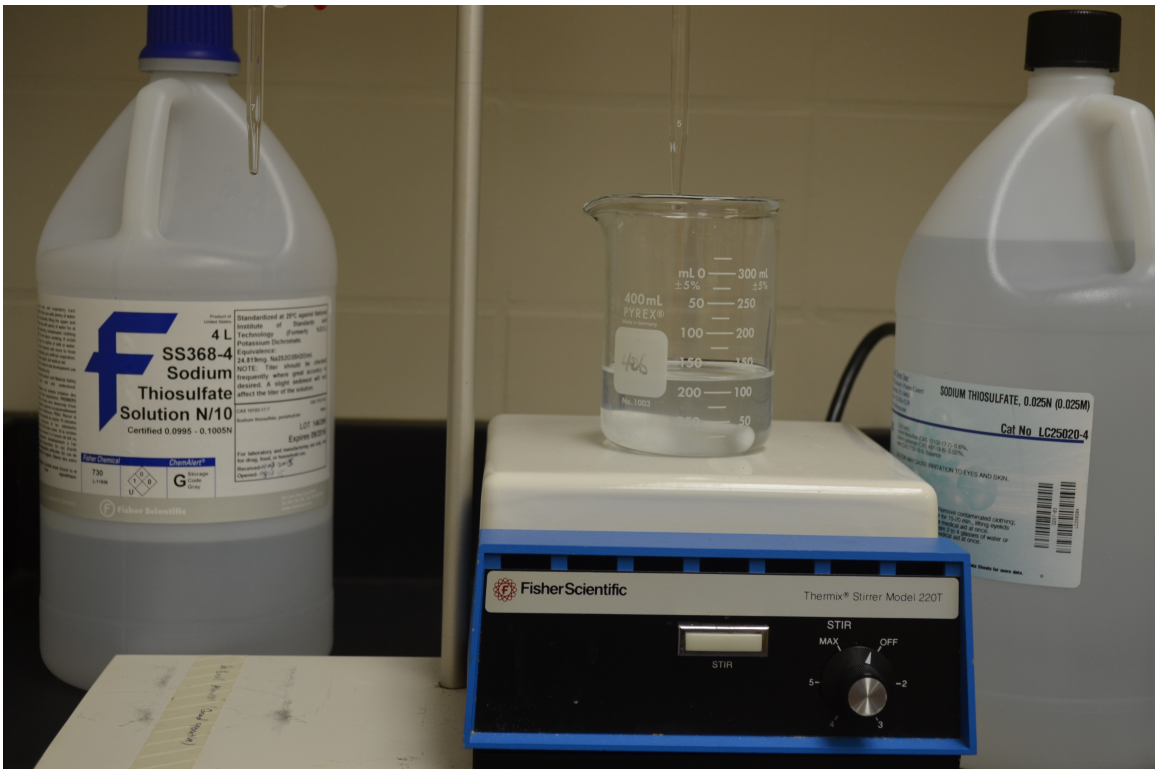

FIGURE 10: The solution becomes colorless after the 2nd titration.

FIGURE 11: Refrigerated pack sent by Evolve Dental containing their product.

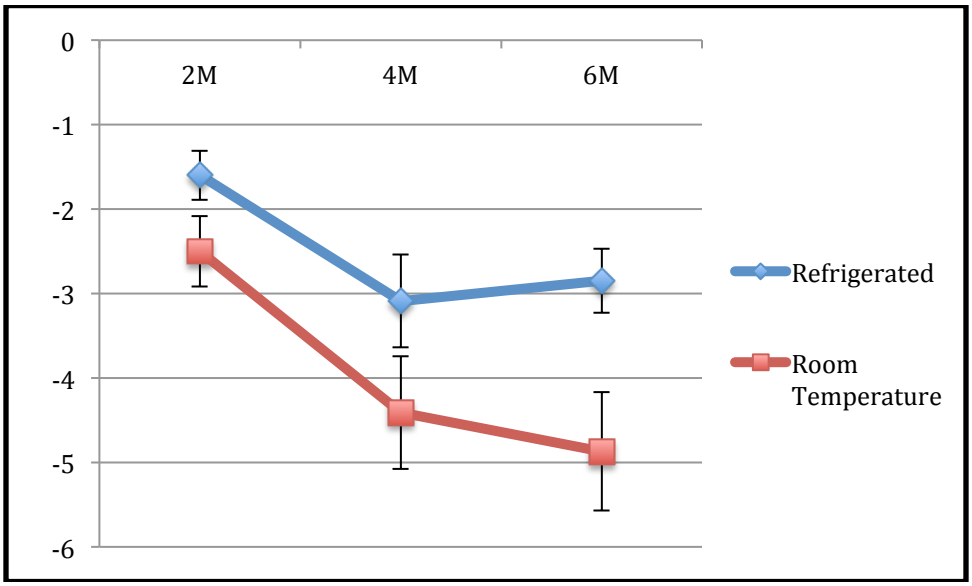

FIGURE 11: Mean for relative change in concentration compared to the baseline concentration (±SE) – graphic.

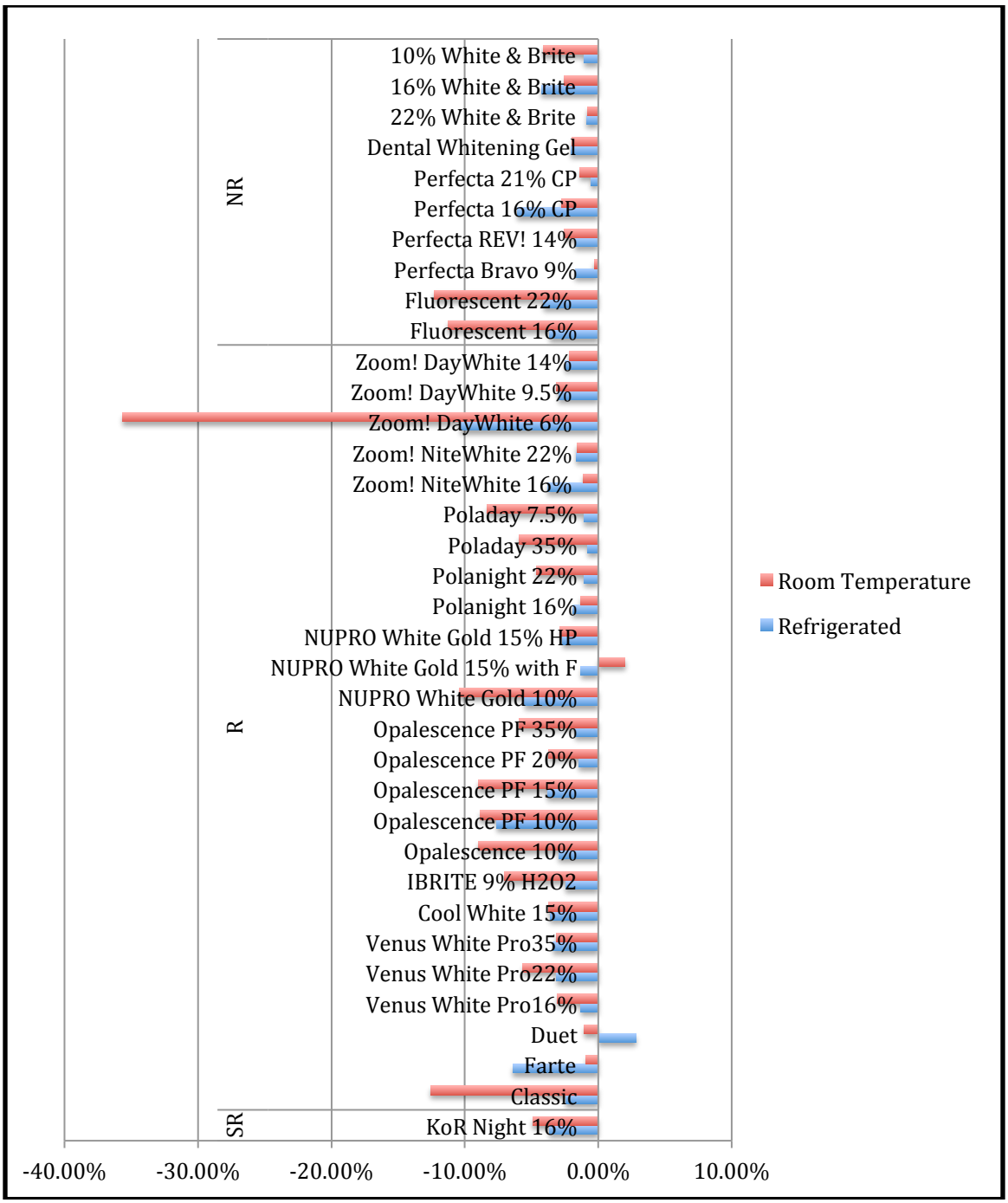

FIGURE 12: Concentrations difference % of each product at 6 months, comparison of refrigerated room temperature samples –graphic.
SR= Refrigeration strongly recommended by the manufacturer.
R= Refrigeration recommended by the manufacturer.
NR= Refrigeration not recommended by the manufacturer.

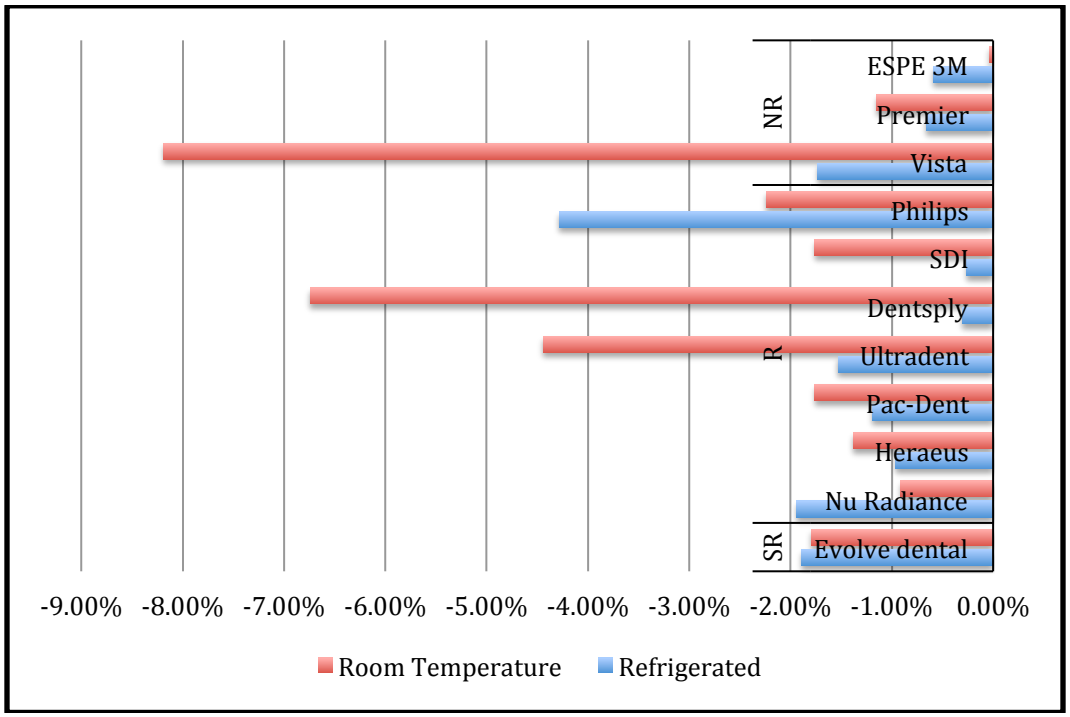

FIGURE 13: Concentrations difference % of each manufacturer at 2 months, comparison of refrigerated to room temperature samples –graphic.
SR= Refrigeration strongly recommended by the manufacturer.
R= Refrigeration recommended by the manufacturer.
NR= Refrigeration not recommended by the manufacturer.

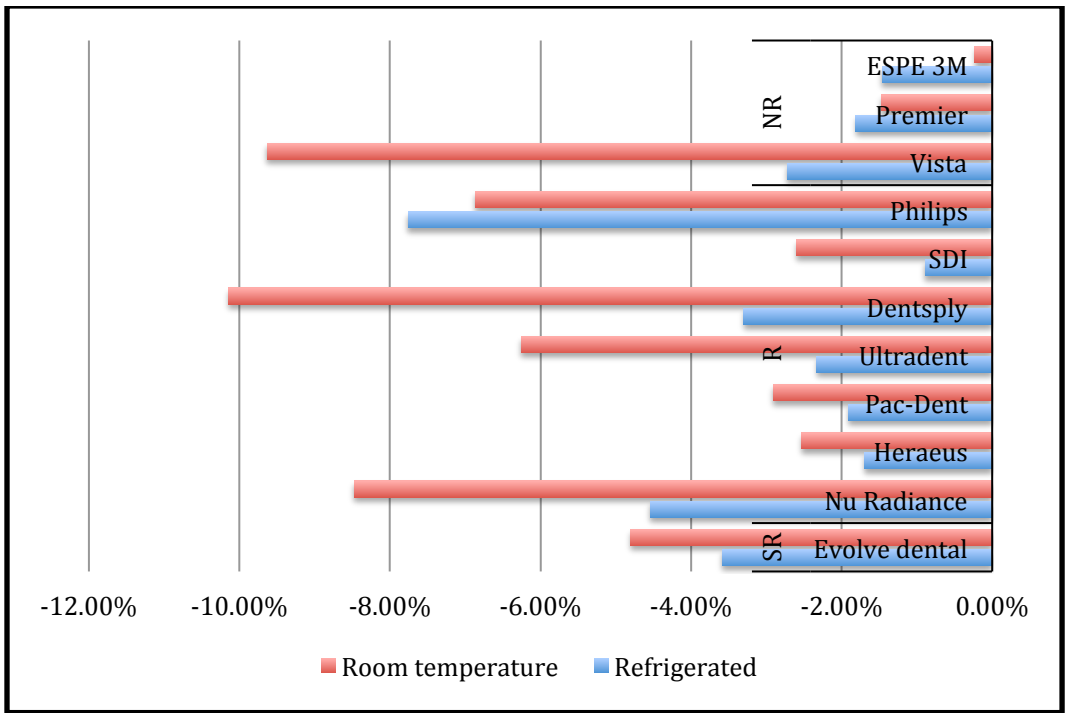

FIGURE 14: Concentrations difference % of each manufacturer at 4 months, comparison of refrigerated to room temperature samples –graphic.
SR= Refrigeration strongly recommended by the manufacturer.
R= Refrigeration recommended by the manufacturer.
NR= Refrigeration not recommended by the manufacturer.

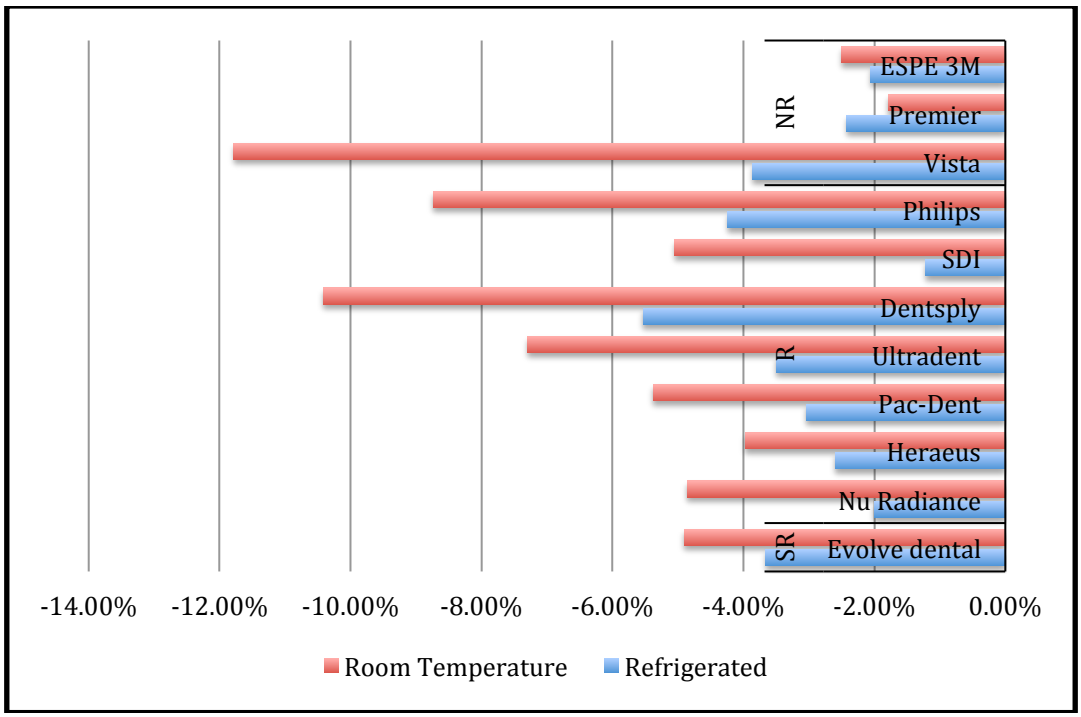

FIGURE 15: Concentrations difference % of each manufacturer at 6 months, comparison of refrigerated to room temperature samples –graphic.
SR= Refrigeration strongly recommended by the manufacturer.
R= Refrigeration recommended by the manufacturer.
NR= Refrigeration not recommended by the manufacturer.

Table I

Products used in this study, lot number, type and concentrations

Manufacturer	Product	Lot #	Type	Conc (Labeled)
Nu Radiance	Classic	140814-1107	CP	22.0%
	Farte	141016-1426	CP	27.0%
	Duet	140827-1120	CP	18.0%
Heraeus	Venus White Pro	1842-B9ZHL	CP	16.0%
	Venus White Pro	1843-B9YJS	CP	22.0%
	Venus White Pro	1940-B9Z99	CP	35.0%
Pac-Dent	Cool White	91514	CP	15.0%
	IBRITE	110714	HP	9.0%
Vista	Fluorescent	2014-2297	CP	16.0%
	Fluorescent	2014-0542	CP	22.0%
Ultradent	Opalescence	B9WYH	CP	10.0%
	Opalescence PF	BB5ZG	CP	10.0%
	Opalescence PF	B9Y9Q	CP	15.0%
	Opalescence PF	BB4TB	CP	20.0%
	Opalescence PF	BBIV8	CP	35.0%
Dentsply	NUPRO White Gold	140714	CP	10.0%
	NUPRO White Gold with F	140728	CP	15.0%
	NUPRO White Gold	140908	HP	15.0%
Premier	Perfecta Bravo	5004	HP	9.0%
	Perfecta REV!	5006	HP	14.0%
	Perfecta	B9PSH	CP	16.0%
	Perfecta	B9X2Z	CP	21.0%
	Dental Whitening Gel Formula	113364	CP	11.0%
Evolve dental	KoR - Night	BB5FM	CP	16.0%
ESPE 3M	White &Brite	N641950	CP	22.0%
	White &Brite	N612564	CP	16.0%
	White &Brite	N6325663	CP	10.0%
SDI	Polanight	P140210	CP	16.0%
	Polanight	P140424	CP	22.0%
	Poladay	P140505	CP	35.0%
	Poladay	P14101Z	HP	7.5%
Philips	Zoom! NiteWhite ACP	14337022	CP	16.0%
	Zoom! NiteWhite ACP	14364002	CP	22.0%
	Zoom! DayWhite ACP	14365025	HP	6.0%
	Zoom! DayWhite ACP	14364014	HP	9.5%
	Zoom! DayWhite ACP	15014017	HP	14.0%

Table II

Storage instructions as recommended by manufacturers

Manufacturer	Product	Storage Instruction
Nu Radiance	Classic Farte Duet	Store at temp not exceeding 73° F (23° C), Protect from direct light especially sunlight and heat sources. Refrigerated for long-term storage. Do not freeze.
Heraeus	Venus White Pro 16% Venus White Pro 22%	Should be refrigerated for long-term storage. Shelf life is 2 years refrigerated.
Heraeus	Venus White Pro 35%	Must be refrigerated for long-term storage. Shelf life is 1 year unrefrigerated and 2 years refrigerated.
Pac-Dent	Cool White 15% CP IBRITE 9% H2O2	Store at or bellow 75°F (24°C). Do not freeze. Shelf life is 24 months under refrigeration and 12 months in cool dry places.
Vista	Fluorescent 16% Fluorescent 22%	Keep out of direct light and heat. Do not freeze.
Ultradent	Opalescence 10% Opalescence PF 10% Opalescence PF 15% Opalescence PF 20% Opalescence PF 35%	Store bleach out of the sun and heat. Refrigeration recommended. Do not freeze.
Dentsply	NUPRO White Gold 10% NUPRO White Gold 15% with F NUPRO White Gold 15% HP	Avoid storage at elevated temperature (86°F/30°C) refrigerate whitening gel when not in use for 2 weeks. Keep away from direct sunlight.

Table II

Continued

Manufacturer	Product	Storage Instruction
Premier	Perfecta Bravo 9% HP Perfecta REV! 14% HP Perfecta 16% CP Perfecta 21% CP Dental Whitening Gel 11%	Store gel at room temperature out of light.
Evolve dental	KoR– Night 16%	Refrigeration recommended from the instant of manufacture until received by the dental practice.
ESPE 3M	22% White & Brite 16% White & Brite 10% White & Brite	Do not freeze. Do not expose to excessive heat or prolonged periods of sunlight.
SDI	Polanight 16% Polanight 22% Poladay 35% Poladay 7.5%	Store gel in the fridge.
Philips	Zoom! NiteWhite 16% ACP Zoom! NiteWhite 22% ACP Zoom! DayWhite 6% ACP Zoom! DayWhite 9.5% ACP Zoom! DayWhite 14% ACP	Should be refrigerated for long-term storage. Shelf life is 2 years refrigerated

Table III

Concentrations of products stored at room temperature (Baseline)

Product	Labeled	BL	D%*
Classic	22.0%	20.76%	-5.65%
Farte	27.0%	25.37%	-6.02%
Duet	18.0%	15.58%	-13.42%
Venus White Pro 16%	16.0%	15.71%	-1.79%
Venus White Pro 22%	22.0%	21.32%	-3.08%
Venus White Pro 35%	35.0%	33.92%	-3.09%
Cool White 15%	15.0%	14.37%	-4.19%
IBRITE 9% H2O2	9.0%	7.38%	**-18.04%**
Fluorescent 16%	16.0%	13.94%	-12.88%
Fluorescent 22%	22.0%	19.63%	-10.78%
Opalescence 10%	10.0%	9.83%	-1.66%
Opalescence PF 10%	10.0%	10.12%	1.18%
Opalescence PF 15%	15.0%	14.52%	-3.23%
Opalescence PF 20%	20.0%	19.35%	-3.23%
Opalescence PF 35%	35.0%	33.95%	-3.01%
NUPRO White Gold 10%	10.0%	8.52%	-14.83%
NUPRO White Gold 15% with F	15.0%	13.99%	-6.72%
NUPRO White Gold 15% HP	15.0%	14.77%	-1.51%
Perfecta Bravo 9%	9.0%	9.29%	3.21%
Perfecta REV! 14%	14.0%	14.69%	4.94%
Perfecta 16% CP	16.0%	13.56%	-15.28%
Perfecta 21% CP	21.0%	18.05%	-14.04%
Dental Whitening Gel	11.0%	10.07%	-8.45%
KoR – Night 16%	16.0%	16.13%	0.82%
22% White & Brite	22.0%	21.19%	-3.66%
16% White & Brite	16.0%	15.35%	-4.08%
10% White & Brite	10.0%	9.38%	-6.23%
Polanight 16%	16.0%	15.02%	-6.11%
Polanight 22%	22.0%	21.08%	-4.19%
Poladay 35%	35.0%	32.65%	-6.72%
Poladay 7.5%	7.5%	7.06%	-5.85%
Zoom! NiteWhite 16% ACP	16.0%	15.20%	-4.99%
Zoom! NiteWhite 22% ACP	22.0%	21.91%	-0.43%
Zoom! DayWhite 6% ACP	6.0%	5.44%	-9.30%
Zoom! DayWhite 9.5% ACP	9.5%	9.36%	-1.45%
Zoom! DayWhite 14% ACP	14.0%	14.16%	1.13%

* Compared to Label concentrations, BL = Baseline

Table IV

Concentrations of products stored at room temperature for 2 months

Product	Labeled	2M	D%*
Classic	22.0%	20.53%	-1.08%
Farte	27.0%	25.01%	-1.43%
Duet	18.0%	15.55%	-0.22%
Venus White Pro 16%	16.0%	15.57%	-0.91%
Venus White Pro 22%	22.0%	20.83%	-2.32%
Venus White Pro 35%	35.0%	33.61%	-0.90%
Cool White 15%	15.0%	14.18%	-1.36%
IBRITE 9% H2O2	9.0%	7.22%	-2.15%
Fluorescent 16%	16.0%	12.68%	-9.05%
Fluorescent 22%	22.0%	18.19%	-7.32%
Opalescence 10%	10.0%	9.34%	-5.02%
Opalescence PF 10%	10.0%	9.32%	-7.90%
Opalescence PF 15%	15.0%	13.87%	-4.46%
Opalescence PF 20%	20.0%	18.76%	-3.05%
Opalescence PF 35%	35.0%	33.34%	-1.78%
NUPRO White Gold 10%	10.0%	7.94%	-6.74%
NUPRO White Gold 15% with F	15.0%	13.00%	-7.11%
NUPRO White Gold 15% HP	15.0%	14.58%	-1.29%
Perfecta Bravo 9%	9.0%	9.28%	-0.05%
Perfecta REV! 14%	14.0%	14.46%	-1.55%
Perfecta 16% CP	16.0%	13.27%	-2.11%
Perfecta 21% CP	21.0%	17.91%	-0.76%
Dental Whitening Gel	11.0%	9.94%	-1.26%
KoR – Night 16%	16.0%	15.84%	-1.79%
22% White & Brite	22.0%	21.17%	-0.11%
16% White & Brite	16.0%	15.35%	-0.01%
10% White & Brite	10.0%	9.38%	-0.01%
Polanight 16%	16.0%	14.98%	-0.30%
Polanight 22%	22.0%	20.84%	-1.12%
Poladay 35%	35.0%	32.31%	-1.05%
Poladay 7.5%	7.5%	6.74%	-4.56%
Zoom! NiteWhite 16% ACP	16.0%	14.89%	-2.05%
Zoom! NiteWhite 22% ACP	22.0%	21.67%	-1.08%
Zoom! DayWhite 6% ACP	6.0%	5.44%	-0.07%
Zoom! DayWhite 9.5% ACP	9.5%	9.06%	-3.19%
Zoom! DayWhite 14% ACP	14.0%	13.47%	-4.82%

* Compared to Label concentrations, 2M = 2 Months

Table V

Concentrations of products stored at room temperature for 4 months

Product	Labeled	4M	D%*
Classic	22.0%	18.52%	-10.79%
Farte	27.0%	24.20%	-4.61%
Duet	18.0%	14.02%	-10.01%
Venus White Pro 16%	16.0%	15.39%	-2.08%
Venus White Pro 22%	22.0%	20.45%	-4.08%
Venus White Pro 35%	35.0%	33.43%	-1.43%
Cool White 15%	15.0%	13.99%	-2.68%
IBRITE 9% H2O2	9.0%	7.15%	-3.13%
Fluorescent 16%	16.0%	12.49%	-10.41%
Fluorescent 22%	22.0%	17.89%	-8.84%
Opalescence 10%	10.0%	9.00%	-8.46%
Opalescence PF 10%	10.0%	9.28%	-8.30%
Opalescence PF 15%	15.0%	13.58%	-6.44%
Opalescence PF 20%	20.0%	18.75%	-3.12%
Opalescence PF 35%	35.0%	32.26%	-4.95%
NUPRO White Gold 10%	10.0%	7.65%	-10.14%
NUPRO White Gold 15% with F	15.0%	12.64%	-9.69%
NUPRO White Gold 15% HP	15.0%	14.35%	-2.84%
Perfecta Bravo 9%	9.0%	9.27%	-0.25%
Perfecta REV! 14%	14.0%	14.45%	-1.66%
Perfecta 16% CP	16.0%	13.26%	-2.16%
Perfecta 21% CP	21.0%	17.81%	-1.36%
Dental Whitening Gel	11.0%	9.88%	-1.92%
KoR – Night 16%	16.0%	15.36%	-4.80%
22% White & Brite	22.0%	21.11%	-0.41%
16% White & Brite	16.0%	15.34%	-0.04%
10% White & Brite	10.0%	9.35%	-0.24%
Polanight 16%	16.0%	14.88%	-0.97%
Polanight 22%	22.0%	20.76%	-1.51%
Poladay 35%	35.0%	31.59%	-3.23%
Poladay 7.5%	7.5%	6.73%	-4.68%
Zoom! NiteWhite 16% ACP	16.0%	15.29%	0.60%
Zoom! NiteWhite 22% ACP	22.0%	21.61%	-1.34%
Zoom! DayWhite 6% ACP	6.0%	5.44%	-0.09%
Zoom! DayWhite 9.5% ACP	9.5%	6.81%	-27.22%
Zoom! DayWhite 14% ACP	14.0%	13.27%	-6.30%

* Compared to baseline concentrations, 4M = 4 Months

Table VI

Concentrations of products stored at room temperature for 6 months

Product	Labeled	6M	D%*
Classic	22.0%	18.15%	-12.57%
Farte	27.0%	25.14%	-0.92%
Duet	18.0%	15.42%	-1.06%
Venus White Pro 16%	16.0%	15.23%	-3.10%
Venus White Pro 22%	22.0%	20.12%	-5.65%
Venus White Pro 35%	35.0%	32.85%	-3.16%
Cool White 15%	15.0%	13.83%	-3.73%
IBRITE 9% H2O2	9.0%	6.86%	-7.02%
Fluorescent 16%	16.0%	12.37%	-11.26%
Fluorescent 22%	22.0%	17.22%	-12.29%
Opalescence 10%	10.0%	8.95%	-9.01%
Opalescence PF 10%	10.0%	9.22%	-8.84%
Opalescence PF 15%	15.0%	13.21%	-9.01%
Opalescence PF 20%	20.0%	18.64%	-3.71%
Opalescence PF 35%	35.0%	31.94%	-5.90%
NUPRO White Gold 10%	10.0%	7.63%	-10.42%
NUPRO White Gold 15% with F	15.0%	14.27%	2.01%
NUPRO White Gold 15% HP	15.0%	14.35%	-2.87%
Perfecta Bravo 9%	9.0%	9.26%	-0.31%
Perfecta REV! 14%	14.0%	14.32%	-2.55%
Perfecta 16% CP	16.0%	13.18%	-2.74%
Perfecta 21% CP	21.0%	17.80%	-1.39%
Dental Whitening Gel	11.0%	9.87%	-1.95%
KoR – Night 16%	16.0%	15.34%	-4.90%
22% White & Brite	22.0%	21.02%	-0.83%
16% White & Brite	16.0%	14.96%	-2.55%
10% White & Brite	10.0%	8.99%	-4.13%
Polanight 16%	16.0%	14.82%	-1.33%
Polanight 22%	22.0%	20.10%	-4.62%
Poladay 35%	35.0%	30.71%	-5.94%
Poladay 7.5%	7.5%	6.48%	-8.31%
Zoom! NiteWhite 16% ACP	16.0%	15.03%	-1.12%
Zoom! NiteWhite 22% ACP	22.0%	21.56%	-1.59%
Zoom! DayWhite 6% ACP	6.0%	3.50%	-35.66%
Zoom! DayWhite 9.5% ACP	9.5%	9.07%	-3.12%
Zoom! DayWhite 14% ACP	14.0%	13.85%	-2.15%

* Compared to baseline concentrations, 6M = 6 Months

Table VII

Concentration of products stored in refrigerator (Baseline)

Product	Labeled	BL	D%*
Classic	22.0%	20.75%	-5.68%
Farte	27.0%	25.44%	-5.76%
Duet	18.0%	15.04%	**-16.42%**
Venus White Pro 16%	16.0%	15.89%	-0.68%
Venus White Pro 22%	22.0%	21.39%	-2.76%
Venus White Pro 35%	35.0%	33.94%	-3.04%
Cool White 15%	15.0%	14.30%	-4.65%
IBRITE 9% H2O2	9.0%	7.42%	**-17.54%**
Fluorescent 16%	16.0%	13.69%	-14.43%
Fluorescent 22%	22.0%	19.19%	-12.76%
Opalescence 10%	10.0%	9.85%	-1.50%
Opalescence PF 10%	10.0%	10.19%	1.93%
Opalescence PF 15%	15.0%	14.95%	-0.36%
Opalescence PF 20%	20.0%	19.48%	-2.59%
Opalescence PF 35%	35.0%	34.12%	-2.52%
NUPRO White Gold 10%	10.0%	8.91%	-10.86%
NUPRO White Gold 15% with F	15.0%	14.54%	-3.05%
NUPRO White Gold 15% HP	15.0%	14.74%	-1.77%
Perfecta Bravo 9%	9.0%	9.63%	7.05%
Perfecta REV! 14%	14.0%	14.84%	6.03%
Perfecta 16% CP	16.0%	14.47%	-9.54%
Perfecta 21% CP	21.0%	18.82%	-10.37%
Dental Whitening Gel	11.0%	10.45%	-5.03%
KoR – Night 16%	16.0%	16.24%	1.53%
22% White & Brite	22.0%	21.07%	-4.21%
16% White & Brite	16.0%	15.34%	-4.12%
10% White & Brite	10.0%	9.55%	-4.48%
Polanight 16%	16.0%	15.23%	-4.80%
Polanight 22%	22.0%	21.15%	-3.86%
Poladay 35%	35.0%	32.68%	-6.63%
Poladay 7.5%	7.5%	6.90%	-7.97%
Zoom! NiteWhite 16% ACP	16.0%	15.59%	-2.55%
Zoom! NiteWhite 22% ACP	22.0%	22.50%	2.29%
Zoom! DayWhite 6% ACP	6.0%	5.88%	-2.03%
Zoom! DayWhite 9.5% ACP	9.5%	9.37%	-1.42%
Zoom! DayWhite 14% ACP	14.0%	14.17%	1.23%

* Compared to label concentrations, BL = Baseline

Table VIII

Concentration of products stored in refrigerator for 2 months

Product	Labeled	2M	D%*
Classic	22.0%	20.48%	-1.30%
Farte	27.0%	24.86%	-2.32%
Duet	18.0%	14.71%	-2.21%
Venus White Pro 16%	16.0%	15.81%	-0.51%
Venus White Pro 22%	22.0%	20.98%	-1.94%
Venus White Pro 35%	35.0%	33.79%	-0.43%
Cool White 15%	15.0%	14.25%	-0.36%
IBRITE 9% H2O2	9.0%	7.27%	-2.01%
Fluorescent 16%	16.0%	13.51%	-1.36%
Fluorescent 22%	22.0%	18.79%	-2.09%
Opalescence 10%	10.0%	9.73%	-1.20%
Opalescence PF 10%	10.0%	9.94%	-2.53%
Opalescence PF 15%	15.0%	14.56%	-2.58%
Opalescence PF 20%	20.0%	19.43%	-0.25%
Opalescence PF 35%	35.0%	33.75%	-1.09%
NUPRO White Gold 10%	10.0%	8.89%	-0.30%
NUPRO White Gold 15% with F	15.0%	13.63%	-6.24%
NUPRO White Gold 15% HP	15.0%	14.66%	-0.50%
Perfecta Bravo 9%	9.0%	9.62%	-0.13%
Perfecta REV! 14%	14.0%	14.82%	-0.17%
Perfecta 16% CP	16.0%	14.10%	-2.58%
Perfecta 21% CP	21.0%	18.80%	-0.13%
Dental Whitening Gel	11.0%	10.42%	-0.29%
KoR – Night 16%	16.0%	15.94%	-1.89%
22% White & Brite	22.0%	20.99%	-0.42%
16% White & Brite	16.0%	15.23%	-0.72%
10% White & Brite	10.0%	9.49%	-0.62%
Polanight 16%	16.0%	15.17%	-0.44%
Polanight 22%	22.0%	21.12%	-0.13%
Poladay 35%	35.0%	32.62%	-0.19%
Poladay 7.5%	7.5%	6.88%	-0.26%
Zoom! NiteWhite 16% ACP	16.0%	15.09%	-3.24%
Zoom! NiteWhite 22% ACP	22.0%	21.67%	-3.71%
Zoom! DayWhite 6% ACP	6.0%	5.40%	-8.15%
Zoom! DayWhite 9.5% ACP	9.5%	8.83%	-5.76%
Zoom! DayWhite 14% ACP	14.0%	14.10%	-0.52%

* Compared to baseline concentrations, 2M = 2 Months

Table IX

Concentration of products stored in refrigerator for 4 months

Product	Labeled	4M	D%*
Classic	22.0%	20.45%	-1.46%
Farte	27.0%	24.42%	-4.04%
Duet	18.0%	13.82%	-8.12%
Venus White Pro 16%	16.0%	15.69%	-1.24%
Venus White Pro 22%	22.0%	20.80%	-2.77%
Venus White Pro 35%	35.0%	33.57%	-1.07%
Cool White 15%	15.0%	14.05%	-1.78%
IBRITE 9% H2O2	9.0%	7.27%	-2.04%
Fluorescent 16%	16.0%	13.42%	-2.00%
Fluorescent 22%	22.0%	18.53%	-3.44%
Opalescence 10%	10.0%	9.59%	-2.68%
Opalescence PF 10%	10.0%	9.84%	-3.46%
Opalescence PF 15%	15.0%	14.44%	-3.37%
Opalescence PF 20%	20.0%	19.34%	-0.74%
Opalescence PF 35%	35.0%	33.65%	-1.38%
NUPRO White Gold 10%	10.0%	8.62%	-3.30%
NUPRO White Gold 15% with F	15.0%	13.61%	-6.44%
NUPRO White Gold 15% HP	15.0%	14.38%	-2.43%
Perfecta Bravo 9%	9.0%	9.56%	-0.76%
Perfecta REV! 14%	14.0%	14.68%	-1.12%
Perfecta 16% CP	16.0%	13.60%	-6.04%
Perfecta 21% CP	21.0%	18.76%	-0.33%
Dental Whitening Gel	11.0%	10.36%	-0.81%
KoR – Night 16%	16.0%	15.66%	-3.58%
22% White & Brite	22.0%	20.93%	-0.68%
16% White & Brite	16.0%	14.89%	-2.92%
10% White & Brite	10.0%	9.48%	-0.78%
Polanight 16%	16.0%	14.94%	-1.94%
Polanight 22%	22.0%	21.03%	-0.59%
Poladay 35%	35.0%	32.57%	-0.34%
Poladay 7.5%	7.5%	6.85%	-0.69%
Zoom! NiteWhite 16% ACP	16.0%	15.01%	-3.74%
Zoom! NiteWhite 22% ACP	22.0%	21.30%	-5.35%
Zoom! DayWhite 6% ACP	6.0%	4.99%	-15.15%
Zoom! DayWhite 9.5% ACP	9.5%	8.12%	-13.25%
Zoom! DayWhite 14% ACP	14.0%	13.99%	-1.31%

* Compared to baseline concentrations, 4M = 4 Months

Table X

Products concentration stored in refrigerator for 6 months

Product	Labeled	6M Conc	D%*
Classic	22.0%	20.24%	-2.44%
Farte	27.0%	23.82%	-6.39%
Duet	18.0%	15.47%	2.84%
Venus White Pro 16%	16.0%	15.69%	-1.30%
Venus White Pro 22%	22.0%	20.73%	-3.12%
Venus White Pro35%	35.0%	32.79%	-3.38%
Cool White 15%	15.0%	13.79%	-3.62%
IBRITE 9% H2O2	9.0%	7.24%	-2.45%
Fluorescent 16%	16.0%	13.19%	-3.63%
Fluorescent 22%	22.0%	18.41%	-4.08%
Opalescence 10%	10.0%	9.56%	-2.95%
Opalescence PF 10%	10.0%	9.42%	-7.60%
Opalescence PF 15%	15.0%	14.38%	-3.79%
Opalescence PF 20%	20.0%	19.20%	-1.44%
Opalescence PF 35%	35.0%	33.54%	-1.70%
NUPRO White Gold 10%	10.0%	8.42%	-5.53%
NUPRO White Gold 15% with F	15.0%	14.36%	-1.29%
NUPRO White Gold 15% HP	15.0%	14.34%	-2.67%
Perfecta Bravo 9%	9.0%	9.47%	-1.71%
Perfecta REV! 14%	14.0%	14.59%	-1.70%
Perfecta 16% CP	16.0%	13.59%	-6.09%
Perfecta 21% CP	21.0%	18.72%	-0.53%
Dental Whitening Gel	11.0%	10.23%	-2.06%
KoR – Night 16%	16.0%	15.65%	-3.66%
22% White & Brite	22.0%	20.89%	-0.88%
16% White & Brite	16.0%	14.69%	-4.24%
10% White & Brite	10.0%	9.45%	-1.05%
Polanight 16%	16.0%	14.94%	-1.93%
Polanight 22%	22.0%	20.93%	-1.06%
Poladay 35%	35.0%	32.41%	-0.81%
Poladay 7.5%	7.5%	6.83%	-1.07%
Zoom! NiteWhite 16% ACP	16.0%	15.00%	-3.78%
Zoom! NiteWhite 22% ACP	22.0%	22.13%	-1.66%
Zoom! DayWhite 6% ACP	6.0%	5.27%	-10.32%
Zoom! DayWhite 9.5% ACP	9.5%	9.09%	-2.95%
Zoom! DayWhite 14% ACP	14.0%	13.82%	-2.49%

* Compared to baseline concentrations, 6M = 6 Months

Table XI

The products concentrations difference % at every time period, sorted by manufacturers' storage recommendation

Fridge	Product	Concentration D%					
		2M		4M		6M	
		Refrigerated	Room Temperature	Refrigerated	Room Temperature	Refrigerated	Room Temperature
SR	KoR Night 16%	-1.89%	-1.79%	-3.58%	-4.80%	-3.66%	-4.90%
R	Classic	-1.30%	-1.08%	-1.46%	-10.79%	-2.44%	-12.57%
	Farte	-2.32%	-1.43%	-4.04%	-4.61%	-6.39%	**-0.92%**
	Duet	-2.21%	-0.22%	-8.12%	-10.01%	**2.84%**	**-1.06%**
	Venus White Pro16%	-0.51%	-0.91%	-1.24%	-2.08%	-1.30%	-3.10%
	Venus White Pro22%	-1.94%	-2.32%	-2.77%	-4.08%	-3.12%	-5.65%
	Venus White Pro35%	-0.43%	-0.90%	-1.07%	-1.43%	-3.38%	-3.16%
	Cool White 15%	-0.36%	-1.36%	-1.78%	-2.68%	-3.62%	-3.73%
	IBRITE 9% H2O2	-2.01%	-2.15%	-2.04%	-3.13%	-2.45%	-7.02%
	Opalescence 10%	-1.20%	-5.02%	-2.68%	-8.46%	-2.95%	-9.01%
	Opalescence PF 10%	-2.53%	-7.90%	-3.46%	-8.30%	-7.60%	-8.84%
	Opalescence PF 15%	-2.58%	-4.46%	-3.37%	-6.44%	-3.79%	-9.01%
	Opalescence PF 20%	-0.25%	-3.05%	-0.74%	-3.12%	-1.44%	-3.71%
	Opalescence PF 35%	-1.09%	-1.78%	-1.38%	-4.95%	-1.70%	-5.90%
	NUPRO White Gold 10%	-0.30%	-6.74%	-3.30%	-10.14%	-5.53%	-10.42%
	NUPRO White Gold 15% with F	-6.24%	-7.11%	-6.44%	-9.69%	**-1.29%**	**2.01%**
	NUPRO White Gold 15% HP	-0.50%	-1.29%	-2.43%	-2.84%	-2.67%	-2.87%
	Polanight 16%	-0.44%	-0.30%	-1.94%	-0.97%	-1.93%	-1.33%
	Polanight 22%	-0.13%	-1.12%	-0.59%	-1.51%	-1.06%	-4.62%
	Poladay 35%	-0.19%	-1.05%	-0.34%	-3.23%	-0.81%	-5.94%
	Poladay 7.5%	-0.26%	-4.56%	-0.69%	-4.68%	-1.07%	-8.31%
	Zoom! NiteWhite 16%	-3.24%	-2.05%	-3.74%	**0.60%**	-3.78%	**-1.12%**
	Zoom! NiteWhite 22%	-3.71%	-1.08%	-5.35%	-1.34%	**-1.66%**	-1.59%
	Zoom! DayWhite 6%	-8.15%	-0.07%	-15.15%	-0.09%	**-10.32%**	-35.66%
	Zoom! DayWhite 9.5%	-5.76%	-3.19%	-13.25%	-27.22%	**-2.95%**	**-3.12%**
	Zoom! DayWhite 14%	-0.52%	-4.82%	-1.31%	-6.30%	-2.49%	**-2.15%**

SR= Refrigeration strongly recommended by the manufacturer.
R= Refrigeration recommended by the manufacturer.
NR= Refrigeration not recommended by the manufacturer.

Table XI

Continuation

Fridge	Product	Concentration D%					
		2M		4M		6M	
		Refrigerated	Room Temperature	Refrigerated	Refrigerated	Refrigerated	Room Temperature
NR	Fluorescent 16%	-1.36%	-9.05%	-2.00%	-10.41%	-3.63%	-11.26%
	Fluorescent 22%	-2.09%	-7.32%	-3.44%	-8.84%	-4.08%	-12.29%
	Perfecta Bravo 9%	-0.13%	-0.05%	-0.76%	-0.25%	-1.71%	-0.31%
	Perfecta REV! 14%	-0.17%	-1.55%	-1.12%	-1.66%	-1.70%	-2.55%
	Perfecta 16% CP	-2.58%	-2.11%	-6.04%	-2.16%	-6.09%	-2.74%
	Perfecta 21% CP	-0.13%	-0.76%	-0.33%	-1.36%	-0.53%	-1.39%
	Dental Whitening Gel	-0.29%	-1.26%	-0.81%	-1.92%	-2.06%	-1.95%
	22% White & Brite	-0.42%	-0.11%	-0.68%	-0.41%	-0.88%	-0.83%
	16% White & Brite	-0.72%	-0.01%	-2.92%	-0.04%	-4.24%	-2.55%
	10% White & Brite	-0.62%	-0.01%	-0.78%	-0.24%	-1.05%	-4.13%

SR= Refrigeration strongly recommended by the manufacturer.
R= Refrigeration recommended by the manufacturer.
NR= Refrigeration not recommended by the manufacturer.

Table XII

The means of products concentrations difference % for each manufacturer at every time period, sorted by manufacturers' storage recommendation

Refrigeration Recommendation	Product	Concentration D%					
		2M		4M		6M	
		Frig	Room Temp	Frig	Room Temp	Frig	Room Temp
Strongly Recommended	Evolve dental	-1.89%	-1.79%	-3.58%	-4.80%	-3.66%	-4.90%
Recommended	Nu Radiance	-1.94%	-0.91%	-4.54%	-8.47%	-2.00%	-4.85%
	Heraeus	-0.96%	-1.38%	-1.69%	-2.53%	-2.60%	-3.97%
	Pac-Dent	-1.19%	-1.76%	-1.91%	-2.91%	-3.04%	-5.38%
	Ultradent	-1.53%	-4.44%	-2.33%	-6.25%	-3.50%	-7.29%
	Dentsply	-0.30%	-6.74%	-3.30%	-10.14%	-5.53%	-10.42%
	SDI	-0.26%	-1.76%	-0.89%	-2.60%	-1.22%	-5.05%
	Philips	-4.28%	-2.24%	-7.76%	-6.87%	-4.24%	-8.73%
Not Recommended	Vista	-1.73%	-8.19%	-2.72%	-9.63%	-3.86%	-11.78%
	Premier	-0.66%	-1.15%	-1.81%	-1.47%	-2.42%	-1.79%
	ESPE 3M	-0.59%	-0.04%	-1.46%	-0.23%	-2.06%	-2.50%

Table XIII

Summary statistics for relative change in concentration compared to the labeled concentration and compared to the baseline concentration

	Storage Method	Time	N	Mean	SD	SE	95% CI for Mean		Min	Max
Label	Refrigerated	BL [A]	36	-4.26	5.56	0.93	-6.14	-2.38	-17.54	7.05
	Room	BL [B]	36	-5.35	5.40	0.90	-7.18	-3.52	-18.04	4.94
Base-line	Refrigerated	2 M [A, a]	36	-1.60	1.76	0.29	-2.19	-1.00	-7.02	-0.13
		4 M [A, b]	36	-3.09	3.30	0.55	-4.20	-1.97	-15.15	-0.33
		6 M [A, b]	36	-2.85	2.30	0.38	-3.63	-2.07	-10.32	2.84
	Room	2 M [B. a]	36	-2.50	2.50	0.42	-3.35	-1.65	-9.05	-0.01
		4 M [B. b]	36	-4.41	4.04	0.67	-5.78	-3.05	-16.54	0.60
		6 M [B, b]	36	-4.87	4.21	0.70	-6.30	-3.45	-17.28	2.01

Upper case letter represent comparison between storage methods.
Lower case letter represent comparison between times.

Table XIV

Mean (95% CI) for relative change in concentration compared to the labeled concentration and compared to the baseline concentration

	Storage Method	Baseline	2 Month	4 Month	6 Month
Label	Refrigerated	-4.3 (-6.1, -2.4) [A]	-	-	-
	Room	-5.4 (-7.2, -3.5) [B]	-	-	-
Baseline	Refrigerated	-	-1.6 (-2.2, -1.0) [A, a]	-3.1 (-4.2, -2.0) [A, b]	-2.8 (-3.6, -2.1) [A, b]
	Room	-	-2.5 (-3.3, -1.7) [B. a]	-4.4 (-5.8, -3.0) [B. b]	-4.9 (-6.3, -3.4) [B. b]

Upper case letter represent comparison between storage methods.
Lower case letter represent comparison between times.

DISCUSSION

The aim of this study was to determine if there is any change in the active agent in the tooth-whitening when it is received from the manufacturer (Baseline), two months, four months, and six months, after it is received, under 2 different storage conditions.

In this study, the methodology used to determine the concentration of carbamide peroxide and hydrogen peroxide is the one stated in the United States Pharmacopeia.[78] It has been widely used in previous bleaching degradation studies.[68, 69, 71, 74, 77] The storage conditions used in this study simulated the storage of at-home bleaching in regular dental clinics and research laboratories. Therefore, room temperature was monitored in the morning during the period the products were stored. The refrigerator temperature was held constant at 5°C (41°F) and the room temperature was within the range from 20.3°C (68.6°F) to 22.78°C (73°F), which were within the range of room temperatures recommended by the bleaching manufacturers.

Statistical analysis revealed a significant decrease in concentration compared to the labeled or baseline concentration for each storage-time combination. Therefore, degradation always happens to the bleaching products after dental practitioners receive them. The manufacturers should strive to keep their products remain the same as the concentration indicated on the label. The results also showed that no significant interaction was found between time and storage method for analyses using the baseline concentration as the reference.

The results obtained showed that the storage method had a significant effect on relative degradation, with more degradation for room storage than for refrigerated storage. The influence of temperature in degradation of carbamide peroxide and hydrogen peroxide was observed from this result. This means that how the bleaching products are stored can influence the clinical response due to degradation of the active agent during storage of the bleaching products in a dental office.

The results also showed that baseline had less degradation than 2 month, 4 month and 6 month. This means that degradation of the bleaching agent is more negatively influenced by the storage condition than by the effect before receiving the products. Less time is required for shipping a bleaching product than for storing it in a dental office.

In addition, the results showed that 2 month had less degradation than 4 month and 6 month, but 4 month and 6 month were not significantly different from each other. One factor that could contribute to making the products degrade more during the storage period might be because the stabilizing agent used by the manufacturers starts to lose its potency gradually until the fourth month of storage. At that point, the stabilizing agent loses potency at slower rate, which makes the bleaching products degrade less.

In this study, 34 products were within 15% of the active agent concentration indicated by the manufacturers at baseline. Six products (Venus White Pro 16%, Opalecence 10%, NUPRO White Gold 15% HP, KoR Night 16%, Zoom! DayWhite 9.5% and Zoom! DayWhite 14%) showed the least concentration difference percentage compared to the labeled concentration. Two products had a 15% lower concentration of the active agent but not more than 30% of that indicated by the manufacturers. These data are comparable to Matis et al. [77] findings in the United States part of their study. They

found that only three out of 35 products had a 15% lower concentration of the active agent but were within the concentration required by the International Organization of Standardization.

In addition, more products had lower than 15% concentrations of what was stated on the label in the following months: four products at the two-month assays, nine products at the four-month assays, and six products at the six-month assays for room-temperature samples. However, fewer products degraded to the level of 15% concentration for the refrigerated samples: three products at the two-month assays, five products at the four-month assays, and three products at the six-month assays (Figure 12) (table XI).

All the bleaching syringes for a specific product were supposed to have the same concentrations since they were from the same lot in this study. Some of the products needed more than one syringe to perform this study during the 6 months period. We noticed an increase in the concentration in the six-month assays in eight products: six of the room-temperature samples (Duet 18%; Nu Radiance, Farte 27%; Nu Radiance, NUPRO White Gold 15% with F; Dentsply, Zoom! NiteWhite 16% ACP; Philips, Zoom! DayWhite 9.5% ACP; Philips and Zoom! DayWhite 14% ACP; Philips) and five of the refrigerated samples (Duet 18%; Nu Radiance, NUPRO White Gold 15% with F; Dentsply, Zoom! NiteWhite 22% ACP, Philips Zoom! DayWhite 6% ACP and Philips Zoom! DayWhite 9.5% ACP, Philips) (Figure 12). Six of these products were designed to have one syringe containing a 2-paste system with a special tip to help in mixing the two pastes together. That might be a contributing factor to contamination and activation of the gel inside the bleaching syringe, which might accelerate the degradation and make a

difference in the concentrations between syringes from the same lot number. In addition, it might be related to the packaging challenges of some manufacturers.

One product (Zoom! DayWhite 6% ACP, Philips) failed to meet the International Standard [78] requirement that products not have 30% lower percentage than indicated on the label throughout the lifetime use of the product in the six-month assays for room-temperature samples.

According to the strength of how manufacturers recommended their product to be stored, two manufacturers, (Dentsply) which recommends refrigeration, and (Vista) which does not, showed the worse mean degradation difference percentage in comparing both refrigerated and non-refrigerated mean degradation percentage of all of their products, the non-refrigerated degradation percentages were higher during all the study period. On the other hand, one manufacturer (Philips), which recommends refrigeration, showed the worst degradation of the refrigerated samples during the first 4 months of the study. One manufacturer (SDI), which recommends refrigeration, showed the least mean degradation percentage under refrigeration at all the time periods of the study (Figure 13,14,15)(Table XII).

There are no published studies available concerning the effect of temperature in bleaching agent degradation for extended storage time. That makes it difficult to compare the results of this study with data from the literature.

The results of this study agree with the hypothesis that the labeled and actual concentrations of the bleaching agent are different at baseline and the concentrations of bleaching agent at baseline, 2 months, 4 months and 6 months are different when maintaining the active agents at room temperature compared to refrigerating them.

Thereby, the conditions of storage of the bleaching products can influence the clinical response due to degradation of the active ingredient. The preferable storage condition verified in this study is refrigeration. Therefore, the bleaching products should be stored in a refrigerator to insure adequate clinical response.

Although the bleaching syringes of a specific product from the same lot were supposed to have the same concentrations, different concentrations were observed in some products. For future studies, we recommend that the entire bleaching package be assayed so the concentrations of each syringe will be known in case of multiple syringes of the same product are used.

SUMMARY AND CONCLUSION

This study was conducted in order to determine if there is any change in the active ingredient of tooth-whitening agents when the products are received from the manufacturer (Baseline), two months, four months, and six months after they are received, under two different storage conditions. Thirty-six products were received from multiple manufacturers: eight with hydrogen peroxide and 28 carbamide with peroxide products. All the bleaching syringes for a specific product were from the same lot. Once the products were received, one sample of each product was stored at room temperature and the other sample was stored in a refrigerator. Assays to determine the baseline concentration were performed within the first two weeks of their arrival and again 2 months, 4 months, and 6 months after receiving the products. All samples were analyzed for peroxide content by using the United States Pharmacopeia recommended method.

The results obtained from this study show the following:

1. Bleaching products have different concentrations than what are indicated on the label.
2. Storage of bleaching products for an extended time at room temperature can cause bleaching products to lose some of their potency.
3. The preferable storage condition verified in this study is under refrigeration.

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APPENDIX

Data Recording Sheet (1) (Carbamide Peroxide)

Evaluator: _____

Date: _ _ / _ _ / _ _ _ _

Lot# _____

Exp. Date: _ _ / _ _ / _ _ _ _

Storage Condition: Room Temp Refrigerator

Manufacturer: _____

Product: _____

Label Conc.: _____ %

Test Month: BL 2 4 6

Trial #: 1 2 3

A. Preparation:

1. Weight Amount of product _____ g**(A)**
2. Add MilliQ (or deionized) water to 100 ml
3. Add 20 ml acetic acid
4. Add potassium iodide _____ g
5. Add 3 drops Ammonium Molybdate. (Dark yellow)

B. Titration:

1. **1st Titration**: Add 0.01 N sodium thiosulfate, use the 50 ml burette, until liquid is pale yellow.

   _____ ml - _____ ml ……... _____ ml **(B)**

2. Add starch indicator (dark purple).
3. **2nd Titration**: Carefully add 0.025 N sodium thiosulfate; use the 10 ml burette, until normal color (Clear).

   _____ ml - _____ ml …… _____ ml **(C)**

C. Calculations:

CP1 % = 4.704 * ___(B)___ ml * (0.1 / ___(A)___ g) = _____ %

CP2 % = 4.704 * ___(C)___ ml * (0.025 / ___(A)___ g) = _____ %

CP % = CP 1 % + CP 2 % = _____ % + _____ % = _____ %

Data Recording Sheet (2) (Hydrogen Peroxide)

Evaluator: _____

Date: _ _ / _ _ / _ _ _ _

Lot# _____

Exp. Date: _ _ / _ _ / _ _ _ _

Storage Condition: Room Temp Refrigerator

Manufacturer: _____

Product: _____

Label Conc.: _____ %

Test Month: BL 2 4 6

Trial #: 1 2 3

D. Preparation:

6. Weight Amount of product _____ g**(A)**
7. Add MilliQ (or deionized) water to 100 ml
8. Add 20 ml acetic acid
9. Add potassium iodide _____ g
10. Add 3 drops Ammonium Molybdate. (Dark yellow)

E. Titration:

4. **1st Titration**: Add 0.01 N sodium thiosulfate, use the 50 ml burette, until liquid is pale yellow.

   _____ ml - _____ ml ……... _____ ml **(B)**

5. Add starch indicator (dark purple).
6. **2nd Titration**: Carefully add 0.025 N sodium thiosulfate; use the 10 ml burette, until normal color (Clear).

   _____ ml - _____ ml …… _____ ml **(C)**

F. Calculations:

HP1 % = 4.704 * ___(B)___ ml * (0.1 / ___(A)___ g) = _____ %

HP2 % = 4.704 * ___(C)___ ml * (0.025 / ___(A)___ g) = _____ %

HP % = HP 1 % + HP 2 % = _____ % + _____ % = _____ %

ABSTRACT

DEGRADATION OF BLEACHING AGENTS UNDER TWO DIFFERENT STORAGE CONDITIONS

by

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The purpose of this study was to determine if there is any change in the active agent in tooth-whitening agents when they are received from the manufacturer (Baseline), 2 months, 4 months and 6 months under 2 different storage conditions.

Eleven manufacturers forwarded pairs of their At-Home bleaching products of various concentrations. Eight with hydrogen peroxide and 28 with carbamide peroxide bleaching agents were received. One sample of each product was stored at room temperature and the other sample was stored in a refrigerator. Assays to determine the concentration were performed within the first 2 weeks of their arrival and at 2 months, 4 months and 6 months after receiving the products.

The protocol, recommended by the United States Pharmacopeia, was used to determine the amount of peroxide in the tooth-whitening agents. The Formula $V \times N \times 1.704/W = HP$ was used to determine the concentration of hydrogen peroxide and the formula $V \times N \times 4.704/W = CP$ was used to determine the concentration of hydrogen peroxide in the carbamide peroxide containing products, where “V” is the volume of

sodium thiosulfate (ml), “W” is the weight of sample (gm) and “N” is the normality of sodium thiosulfate.

There was a significant decrease in concentration compared to the labeled concentration and compared to the baseline concentration for each storage-time combination. No significant interaction was found between time and storage method. Storage method had a significant effect on relative degradation, with more degradation for room storage than for refrigerated storage.

Using the baseline concentration as the reference, 2 month had less degradation than 4 month and 6 month, but 4 month and 6 month were not significantly different from each other.

The conclusions of this study are:

1. Bleaching products have different concentrations than what are indicated on the label.
2. Storage of bleaching products for an extended time at room temperature can cause bleaching products to lose some of their potency.
3. The preferable storage condition verified in this study is under refrigeration.

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